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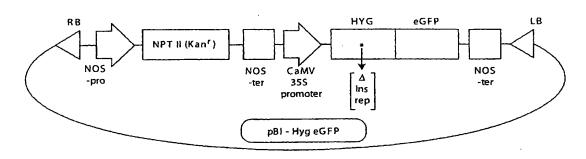
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(54) Title: TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES

11/92512 A2



(57) Abstract: Presented are methods and compositions for targeted chromosomal genomic alterations with modified single-stranded oligonucleotides. The oligonucleotides of the invention have modified nuclease-resisant termini comprising LNA, phosphorothioate linkages or 2'-O-Me base analogues or combinations of such modifications.

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TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES

Field Of The Invention

The technical field of the invention is oligonucleotide-directed repair or alteration of plant genetic information using novel chemically modified oligonucleotides.

Background Of The Invention

A number of methods have been developed specifically to alter the genomic information of plants. These methods generally include the use of vectors such as, for example, T-DNA, carrying nucleic acid sequences encoding partial or complete portions of a particular protein which is expressed in a cell or tissue to effect the alteration. The expression of the particular protein then results in the desired phenotype. See, for example, United States Patent 4,459,355 which describes a method for transforming plants with a DNA vector and United States Patent 5,188,642 which describes cloning or expression vectors containing a transgenic DNA sequence which when expressed in plants confers resistance to the herbicide glyphosate. The use of such transgene-containing vectors adds one or more exogenous copies of a gene in a usually random fashion at one or more integration sites of the plant's genome at some variable frequency. The introduced gene may be foreign or may be derived from the host plant. Any gene which was originally present in the genome, which may be, for example, a normal allelic variant, mutated, defective, and/or functional copy of the introduced gene, is retained in the genome of the host plant.

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These methods of gene alteration are problematic in that complications which can compromise the vigor, productivity, yield, etc. of the plant may result. One such problem is that insertion of exogenous nucleic acid at random location(s) in the genome can have deleterious effects. The random nature of this insertion and/or the use of exogenous promoters can also cause the timing, location or strength of expression of the introduced transgene to be inappropriate or unpredictable. Another problem with such systems includes the addition of unnecessary and unwanted genetic material to the genome of the recipient, including, for example, T-DNA ends or other vector remnants, exogenous control sequences required to allow production of the transgene protein, which control sequences may be

exogenous or native to the host plant and/or the transgene, and reporter genes or resistance markers. Such remnants and added sequences may have presently unrecognized consequences, for example, involving genetic rearrangements of the recipient genomes. In addition, concerns have been raised with consumption, especially by humans, of plants containing such exogenous genetic material.

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More recently, simpler systems involving poly- or oligo- nucleotides have been described for use in the alteration of genomic DNA. These chimeric RNA-DNA oligonucleotides, requiring contiguous RNA and DNA bases in a double-stranded molecule folded by complementarity into a double hairpin conformation, have been shown to effect single basepair or frameshift alterations, for example, for mutation or repair of plant, animal or fungal genomes. See, for example, WO 99/07865 and U.S. Patent 5,565,350. In the chimeric RNA-DNA oligonucleotide, an uninterrupted stretch of DNA bases within the molecule is required for sequence alteration of the targeted genome while the obligate RNA residues are involved in complex stability. Due to the length, backbone composition, and structural configuration of these chimeric RNA-DNA molecules, they are expensive to synthesize and difficult to purify. Moreover, if the RNA-containing strand of the chimeric RNA-DNA oligonucleotide is designed so as to direct gene alteration, a series of mutagenic reactions resulting in nonspecific base alteration can result. Such a result reduces the utility of such a molecule in methods designed for targeted gene alteration.

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Alternatively, other oligo- or poly- nucleotides have been used which require a triplex forming, usually polypurine or polypyrimidine, structural domain which binds to a DNA helical duplex through Hoogsteen interactions between the major groove of the DNA duplex and the oligonucleotide. Such oligonucleotides may have an additional DNA reactive moiety, such as psoralen, covalently linked to the oligonucleotide. These reactive moieties function as effective intercalation agents, stabilize the formation of a triplex and can be mutagenic. Such agents may be required in order to stabilize the triplex forming domain of the oligonucleotide with the DNA double helix if the Hoogsteen interactions from the oligonucleotide/target base composition are insufficient. See, e.g., U.S. Patent 5,422,251. The utility of these oligonucleotides for directing targeted gene alteration is compromised by a high frequency of nonspecific base changes.

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In more recent work, the domain for altering a genome is linked or tethered to the triplex forming domain of the bi-functional oligonucleotide, adding an additional linking or tethering functional domain to the oligonucleotide. See, e.g., Culver et al., Nature Biotechnology 17: 989-93 (1999). Such chimeric or triplex forming molecules have distinct structural requirements for each of the different domains of the complete poly- or oligo-nucleotide in order to effect the desired genomic alteration in either episomal or chromosomal targets.

Other genes, e.g. CFTR, have been targeted by homologous recombination using duplex fragments having several hundred basepairs. See, e.g., Kunzelmann et al., <u>Gene Ther.</u> 3:859-867 (1996). Similar efforts to target genes by homologous recombination in plants using large fragments of DNA had some success. See Kempin et al., <u>Nature</u> 389:802-803 (1997). However, the efficiency and reproducibility of the published homologous recombination approach in plants has severely limited the widespread use of this method.

Earlier experiments to mutagenize an antibiotic resistance indicator gene by homologous recombination used an unmodified DNA oligonucleotide rather than larger fragments of DNA, wherein the oligonucleotide had no functional domains other than a region of complementary sequence to the target. See Campbell et al., New Biologist 1: 223-227 (1989). These experiments required large concentrations of the oligonucleotide, exhibited a very low frequency of episomal modification of a targeted exogenous plasmid gene not normally found in the cell and have not been reproduced. However, as shown in examples herein, we have observed that an unmodified DNA oligonucleotide can convert a base at low frequency which is detectable using the assay systems described herein.

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Oligonucleotides designed for use in the targeted alteration of genetic information are significantly different from oligonucleotides designed for antisense approaches. For example, antisense oligonucleotides are perfectly complementary to and bind an mRNA strand in order to modify expression of a targeted mRNA and are used at high concentration. As a consequence, they are unable to produce a gene conversion event by either mutagenesis or repair of a defect in the chromosomal DNA of a host genome. Furthermore, the backbone chemical composition used in most oligonucleotides designed for use in antisense approaches renders them inactive as substrates for homologous pairing or mismatch repair enzymes and the high concentrations of oligonucleotide required for antisense applications can be toxic with some types of nucleotide modifications. In addition, antisense oligonucleotides must be complementary to the mRNA and therefore, may not be complementary to the other DNA strand or to genomic sequences that span the junction between intron sequence and exon sequence.

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Artificial chromosomes can be useful for the screening purposes identified herein. These molecules are man-made linear or circular DNA molecules constructed from essential cis-acting DNA sequence elements that are responsible for the proper replication and partitioning of natural chromosomes (Murray et al., 1983). The essential elements are: (1) Autonomous Replication Sequences (ARS), (2) Centromeres, and (3) Telomeres.

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Yeast artificial chromosomes (YACs) allow large segments of genomic DNA to be cloned and modified (Burke et al., Science 236:806; Peterson et al., Trends Genet. 13:61 (1997); Choi, et al., Nat.

Genet., 4:117-223 (1993), Davies, et al., Biotechnology 11:911-914 (1993), Matsuura, et al., Hum. Mol. Genet., 5:451-459 (1996), Peterson et al., Proc. Natl. Acad. Sci., 93:6605-6609 (1996); and Schedl, et al., Cell, 86:71-82 (1996)). Other vectors also have been developed for the cloning of large segments of genomic DNA, including cosmids, and bacteriophage P1 (Sternberg et al., Proc. Natl. Acad. Sci. U.S.A., 87:103-107 (1990)). YACs have certain advantages over these alternative large capacity cloning vectors (Burke et al., Science, 236:806-812 (1987)). The maximum insert size is 35-30 kb for cosmids, and 100 kb for bacteriophage P1, both of which are much smaller than the maximal insert size for a YAC.

An alternative to YACs are cloning systems based on the *E. coli* fertility factor that have been developed to construct large genomic DNA insert libraries. They are bacterial artificial chromosomes (BACs) and P-1 derived artificial chromosomes (PACs) (Mejia et al., Genome Res. 7:179-186 (1997); Shizuya et al., Proc. Natl. Acad. Sci. 89:8794-8797 (1992); Ioannou et al., Nat. Genet., 6:84-89 (1994); Hosoda et al., Nucleic Acids Res. 18:3863 (1990)). BACs are based on the *E. coli* fertility plasmid (F factor); and PACs are based on the bacteriophage P1. These vectors propagate at a very low copy number (1-2 per cell) enabling genomic inserts up to 300 kb in size to be stably maintained in recombination deficient hosts. The PACs and BACs are circular DNA molecules that are readily isolated from the host genomic background by classical alkaline lysis (Birnboim et al., Nucleic Acids Res. 7:1513-1523 (1979)). In addition, BACs have been developed for transformation of plants with highmolecular weight DNA using the T-DNA system (Hamilton, Gene 24:107-116 (1997); Frary & Hamilton, Transgenic Res. 10: 121-132 (2001)).

A need exists for simple, inexpensive oligonucleotides capable of producing targeted alteration of genetic material such as those described herein as well as methods to identify optimal oligonucleotides that accurately and efficiently alter target DNA.

Summary Of The Invention

Novel, modified single-stranded nucleic acid molecules that direct gene alteration in plants are identified and the efficiency of alteration is analyzed both *in vitro* using a cell-free extract assay and *in vivo* using a yeast system and a plant system. The alteration in an oligonucleotide of the invention may comprise an insertion, deletion, substitution, as well as any combination of these. Site specific alteration of DNA is not only useful for studying function of proteins *in vivo*, but it is also useful for creating plants with desired phenotypes, including, for example, environmental stress tolerance, improved nutritional value, herbicide resistance, disease resistance, modified oil production, modified starch production, and altered floral morphology including selective sterility. As described herein,

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Oligonucleotides of the invention target directed specific gene alterations in genomic double-stranded DNA in cells. The target genomic DNA can be nuclear chromosomal DNA as well as plastid or mitochondrial chromosomal DNA. The target DNA can also be a transgene present in the plant cell, including; for example, a previously introduced T-DNA. For screening purposes, the target plant DNA can also be extrachromosomal DNA present in plant or non-plant cells in various forms including, e.g., mammalian artificial chromosomes (MACs), PACs from P-1 vectors, yeast artificial chromosomes (YACs), bacterial artificial chromosomes (BACs), plant artificial chromosomes (PLACs), as well as episomal DNA, including episomal DNA from an exogenous source such as a plasmid or recombinant vector. Many of these artificial chromosome constructs containing plant DNA can be obtained from a variety of sources, including, e.g., the Arabidopsis Biological Resource Center (ABRC) at the Ohio State University, and the Rice Genome Research Program at the MAFF DNA bank in Ibaraki, Japan. The target DNA may be transcriptionally silent or active. In a preferred embodiment, the target DNA to be altered is the non-transcribed strand of a genomic DNA duplex. In a more preferred embodiment, the target DNA to be altered is the non-transcribed strand of a transcribed gene of a genomic DNA duplex.

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The low efficiency of targeted gene alteration obtained using unmodified DNA oligonucleotides is believed to be largely the result of degradation by nucleases present in the reaction mixture or the target cell. Although different modifications are known to have different effects on the nuclease resistance of oligonucleotides or stability of duplexes formed by such oligonucleotides (see, e.g., Koshkin et al., J. Am. Chem. Soc., 120:13252-3), we have found that it is not possible to predict which of any particular known modification would be most useful for any given alteration event, including for the construction of gene alteration oligonucleotides, because of the interaction of different as yet unidentified proteins during the gene alteration event. Herein, a variety of nucleic acid analogs have been developed that increase the nuclease resistance of oligonucleotides that contain them, including, e.g., nucleotides containing phosphorothicate linkages or 2'-O-methyl analogs. We recently discovered that single-stranded DNA oligonucleotides modified to contain 2'-O-methyl RNA nucleotides or phosphorothioate linkages can enable specific alteration of genetic information at a higher level than either unmodified single-stranded DNA or a chimeric RNA/DNA molecule. See, for example, copending applications United States application no. 60/208,538, United States application no. 60/244,989, United States application no. 09/818,875, international application no. PCT/US01/09761 and Gamper et al., Nucleic Acids Research 28: 4332-4339 (2000), the disclosures of which are incorporated herein in their entirety by reference. We also found that additional nucleic acid analogs which increase the nuclease resistance of oligonucleotides that contain them, including, e.g., "locked nucleic acids" or "LNAs", xyloWO 01/92512 PCT/US01/17672 - 6 -

LNAs and L-ribo-LNAs; see, for example, Wengel & Nielsen, WO 99/14226; Wengel, WO 00/56748; Wengel, WO 00/66604; and Jakobsen & Koshkin, WO 01/25478 also allow specific targeted alteration of genetic information.

The assay allows for determining the optimum length of the oligonucleotide, optimum sequence of the oligonucleotide, optimum position of the mismatched base or bases, optimum chemical modification or modifications, optimum strand targeted for identifying and selecting the most efficient oligonucleotide for a particular gene alteration event by comparing to a control oligonucleotide. Control oligonucleotides may include a chimeric RNA-DNA double hairpin oligonucleotide directing the same gene alteration event, an oligonucleotide that matches its target completely, an oligonucleotide in which all linkages are phosphorothiolated, an oligonucleotide fully substituted with 2'-O-methyl analogs or an RNA oligonucleotide. Such control oligonucleotides either fail to direct a targeted alteration or do so at a lower efficiency as compared to the oligonucleotides of the invention. The assay further allows for determining the optimum position of a gene alteration event within an oligonucleotide, optimum concentration of the selected oligonucleotide for maximum alteration efficiency by systematically testing a range of concentrations, as well as optimization of either the source of cell extract by testing different plants or strains, or testing cells derived from different plants or strains, or plant cell lines. Using a series of single-stranded oligonucleotides, comprising all RNA or DNA residues and various mixtures of the two, several new structures are identified as viable molecules in nucleotide conversion to direct or repair a genomic mutagenic event. When extracts from mammalian, plant and fungal cells are used and are analyzed using a genetic readout assay in bacteria, single-stranded oligonucleotides having one of several modifications are found to be more active than a control RNA-DNA double hairpin chimera structure when evaluated using an in vitro gene repair assay. Similar results are also observed in vivo using yeast, mammalian and plant cells. Molecules containing various lengths of modified bases were found to possess greater activity than unmodified single-stranded DNA molecules.

Detailed Description Of The Invention

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The present invention provides oligonucleotides having chemically modified, nuclease resistant residues, preferably at or near the termini of the oligonucleotides, and methods for their identification and use in targeted alteration of plant genetic material, including gene mutation, targeted gene repair and gene knockout. The oligonucleotides are preferably used for mismatch repair or alteration by changing at least one nucleic acid base, or for frameshift repair or alteration by addition or deletion of at least one nucleic acid base. The oligonucleotides of the invention direct any such alteration,

including gene correction, gene repair or gene mutation and can be used, for example, to introduce a polymorphism or haplotype or to eliminate ("knockout") a particular protein activity. For example, gene alterations that knockout a particular protein activity can be obtained using oligonucleotides designed to convert a codon in the coding region of the protein to a stop codon, thus prematurely terminating translation of the protein. Oligonucleotides that introduce stop codons in the open-reading-frame of the protein are one embodiment of the invention. Generally, oligonucleotides that introduce stop codons early in the open-reading-frame of the protein are preferred. If the open-reading-frame contains more than one methionine, oligonucleotides that introduce stop codons after the second methionine are preferred. Additionally, if the gene exhibits alternative splice sites, oligonucleotides that introduce stop codons in exons after the alternative splice site are preferred. The following table provides examples of codons that can be converted to stop codons by altering a single oligonucleotide. A skilled artisan could readily identify other codons that can be converted to stop codons by altering one, two or three of the base pairs in a given codon. Similarly, a skilled artisan could readily identify codons that can be converted to stop codons by a frameshift mutations that inserts or deletes one or two base pairs in the open-reading-frame. It is also understood that more than one stop codon can be generated in a single open-reading-frame and that these stop codons can be adjacent in the sequence or separated by intervening codons. Where more than one stop codon is introduced into a single open-reading-frame, such alterations can be generated by a single or multiple oligonucleotides and can be generated simultaneously or by sequential mutagenesis of the target nucleic acid.

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Original codons*	Corresponding stop codon
GGA (glycine), AGA (arginine), CGA (arginine), TTA (leucine),	TGA
TCA (serine), TGT (cysteine), TGG (tryptophan), TGC (cysteine)	
AAG (lysine), GAG (glutamate), CAG (glutamine), TTG (leucine),	TAG
TCG (serine), TGG (tryptophan), TAT (cysteine), TAC (tyrosine)	
AAA (lysine), GAA (glutamate), CAA (glutamine), TTA (leucine),	TAA
TCA (serine), TAT (cysteine), TAC (tyrosine)	

*The amino acid encoded by the original codon is shown in parentheses and the base targeted for alteration to convert the codon to the corresponding stop codon is underlined and in bold

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The oligonucleotides of the invention are designed as substrates for homologous pairing and repair enzymes and as such have a unique backbone composition that differs from chimeric RNA-DNA double hairpin oligonucleotides, antisense oligonucleotides, and/or other poly- or oligo-nucleotides used for altering genomic DNA, such as triplex forming oligonucleotides. The single-stranded oligonucleotides described herein are inexpensive to synthesize and easy to purify. In side-by-side comparisons, an optimized single-stranded oligonucleotide comprising modified residues as described herein is significantly more efficient than a chimeric RNA-DNA double hairpin oligonucleotide in directing a base substitution or frameshift mutation in a cell-free extract assay.

We have discovered that single-stranded oligonucleotides having a DNA domain surrounding the targeted base, with the domain preferably central to the poly- or oligo-nucleotide, and having at least one modified end, preferably at the 3' terminal region, are able to alter a target genetic sequence and with an efficiency that is higher than chimeric RNA-DNA double hairpin oligonucleotides disclosed in US Patent 5,565,350. Preferred oligonucleotides of the invention have at least two modified bases on at least one of the termini, preferably the 3' terminus of the oligonucleotide. Oligonucleotides of the invention can efficiently be used to introduce targeted alterations in a genetic sequence of DNA in the presence of human, animal, plant, fungal (including yeast) proteins and in cells of different types including, for example, plant cells, fungal cells including S. cerevisiae, Ustillago maydis, Candida albicans, and mammalian cells. Particularly preferred are cells and cell extracts derived from plants including, for example, experimental model plants such as Chlamydomonas reinhardtii, Physcomitrella patens, and Arabidopsis thaliana in addition to crop plants such as cauliflower (Brassica oleracea), artichoke (Cvnara scolymus), fruits such as apples (Malus, e.g. domesticus), mangoes (Mangifera, e.g. indica), banana (Musa, e.g. acuminata), berries (such as currant, Ribes, e.g. rubrum), kiwifruit (Actinidia, e.g. chinensis), grapes (Vitis, e.g. vinifera), bell peppers (Capsicum, e.g. annuum), cherries (such as the sweet cherry, Prunus, e.g. avium), cucumber (Cucumis, e.g. sativus), melons (Cucumis, e.g. melo), nuts (such as walnut, Juglans, e.g. regia; peanut, Arachis hypogeae), orange (Citrus, e.g. maxima), peach (Prunus, e.g. persica), pear (Pyra, e.g. communis), plum (Prunus, e.g. domestica), strawberry (Fragaria, e.g. moschata or vesca), tomato (Lycopersicon, e.g. esculentum); leaves and forage, such as alfalfa (Medicago, e.g. sativa or truncatula), cabbage (e.g. Brassica oleracea), endive (Cichoreum, e.g. endivia), leek (Allium, e.g. porrum), lettuce (Lactuca, e.g. sativa), spinach (Spinacia, e.g. oleraceae), tobacco (Nicotiana, e.g. tabacum); roots, such as arrowroot (Maranta, e.g. arundinacea), beet (Beta, e.g. vulgaris), carrot (Daucus, e.g. carota), cassava (Manihot, e.g. esculenta), turnip (Brassica, e.g. rapa), radish (Raphanus, e.g. sativus), yam (Dioscorea, e.g. esculenta), sweet potato (Ipomoea batatas); seeds, including oilseeds,

such as beans (Phaseolus, e.g. vulgaris), pea (Pisum, e.g. sativum), soybean (Glycine, e.g. max), cowpea (Vigna unguiculata), mothbean (Vigna aconitifolia), wheat (Triticum, e.g. aestivum), sorghum (Sorghum e.g. bicolor), barley (Hordeum, e.g. vulgare), corn (Zea, e.g. mays), rice (Oryza, e.g. sativa), rapeseed (Brassica napus), millet (Panicum sp.), sunflower (Helianthus annuus), oats (Avena sativa), chickpea (Cicer, e.g. arietinum); tubers, such as kohlrabi (Brassica, e.g. oleraceae), potato (Solanum, e.g. tuberosum) and the like; fiber and wood plants, such as flax (Linum e.g. usitatissimum), cotton (Gossypium e.g. hirsutum), pine (Pinus sp.), oak (Quercus sp.), eucalyptus (Eucalyptus sp.), and the like and ornamental plants such as turfgrass (Lolium, e.g. rigidum), petunia (Petunia, e.g. x hybrida), hyacinth (Hyacinthus orientalis), carnation (Dianthus e.g. caryophyllus), delphinium (Delphinium, e.g. ajacis), Job's tears (Coix lacryma-jobi), snapdragon (Antirrhinum majus), poppy (Papaver, e.g. nudicaule), lilac (Syringa, e.g. vulgaris), hydrangea (Hydrangea e.g. macrophylla), roses (including Gallicas, Albas, Damasks, Damask Perpetuals, Centifolias, Chinas, Teas and Hybrid Teas) and ornamental goldenrods (e.g. Solidago spp.). Such plant cells can then be used to regenerate whole plants according to methods described herein or any method known in the art. The DNA domain of the oligonucleotides is preferably fully complementary to one strand of the gene target, except for the mismatch base or bases responsible for the gene alteration event(s). On either side of the preferably central DNA domain, the contiguous bases may be either RNA bases or, preferably, are primarily DNA bases. The central DNA domain is generally at least 8 nucleotides in length. The base(s) targeted for alteration in the most preferred embodiments are at least about 8, 9 or 10 bases from one end of the oligonucleotide.

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According to certain embodiments, one or both of the termini of the oligonucleotides of the present invention comprise phosphorothioate modifications, LNA backbone (including LNA derivatives and analogs) modifications, or 2'-O-methyl base analogs, or any combination of these modifications. Oligonucleotides comprising 2'-O-methyl or LNA analogs are a mixed DNA/RNA polymer. The oligonucleotides of the invention are, however, single-stranded and are not designed to form a stable internal duplex structure within the oligonucleotide. The efficiency of gene alteration is surprisingly increased with oligonucleotides having internal complementary sequence comprising phosphorothioate modified bases as compared to 2'-O-methyl modifications. This result indicates that specific chemical interactions are involved between the converting oligonucleotide and the proteins involved in the conversion. The effect of other such chemical interactions to produce nuclease resistant termini using modifications other than LNA (including LNA derivatives or analogs), phosphorothioate linkages, or 2'-O-methyl analog incorporation into an oligonucleotide can not yet be predicted because the proteins

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involved in the alteration process and their particular chemical interaction with the oligonucleotide substituents are not yet known and cannot be predicted.

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In the examples, oligonucleotides of defined sequence are provided for alteration of genes in particular plants. Provided the teachings of the instant application, one of skill in the art could readily design oligonucleotides to introduce analogous alterations in homologous genes from any plant. Furthermore, in the tables of these examples, the oligonucleotides of the invention are not limited to the particular sequences disclosed. The oligonucleotides of the invention include extensions of the appropriate sequence of the longer 120 base oligonucleotides which can be added base by base to the smallest disclosed oligonucleotides of 17 bases. Thus the oligonucleotides of the invention include for each correcting change, oligonucleotides of length 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, or 120 with further single-nucleotide additions up to the longest sequence disclosed. In some embodiments, longer nucleic acids of up to 240 bases which comprise the sequences disclosed herein may be used. Moreover, the oligonucleotides of the invention do not require a symmetrical extension on either side of the central DNA domain. Similarly, the oligonucleotides of the invention as disclosed in the various tables for alteration of particular plant genes contain phosphorothioate linkages, 2'-O-methyl analog or LNA (including LNA derivatives and analogs) or any combination of these modifications just as the assay oligonucleotides do.

The present invention, however, is not limited to oligonucleotides that contain any particular nuclease resistant modification. Oligonucleotides of the invention may be altered with any combination of additional LNAs (including LNA derivatives and analogs), phosphorothioate linkages or 2'-O-methyl analogs to maximize conversion efficiency. For oligonucleotides of the invention that are longer than about 17 to about 25 bases in length, internal as well as terminal region segments of the backbone may be altered. Alternatively, simple fold-back structures at each end of a oligonucleotide or appended end groups may be used in addition to a modified backbone for conferring additional nuclease resistance.

The different oligonucleotides of the present invention preferably contain more than one of the aforementioned backbone modifications at each end. In some embodiments, the backbone modifications are adjacent to one another. However, the optimal number and placement of backbone modifications for any individual oligonucleotide will vary with the length of the oligonucleotide and the particular type of backbone modification(s) that are used. If constructs of identical sequence having

phosphorothioate linkages are compared, 2, 3, 4, 5, or 6 phosphorothioate linkages at each end are preferred. If constructs of identical sequence having 2'-O-methyl base analogs are compared, 1, 2, 3 or 4 analogs are preferred. The optimal number and type of backbone modifications for any particular oligonucleotide useful for altering target DNA may be determined empirically by comparing the alteration efficiency of the oligonucleotide comprising any combination of the modifications to a control molecule of comparable sequence using any of the assays described herein. The optimal position(s) for oligonucleotide modifications for a maximally efficient altering oligonucleotide can be determined by testing the various modifications as compared to control molecule of comparable sequence in one of the assays disclosed herein. In such assays, a control molecule includes, e.g., a completely 2'-O-methyl substituted molecule, a completely complementary oligonucleotide, or a chimeric RNA-DNA double hairpin.

Increasing the number of phosphorothioate linkages, LNAs or 2'-O-methyl bases beyond the preferred number generally decreases the gene repair activity of a 25 nucleotide long oligonucleotide. Based on analysis of the concentration of oligonucleotide present in the extract after different time periods of incubation, it is believed that the terminal modifications impart nuclease resistance to the oligonucleotide thereby allowing it to survive within the cellular environment. However, this may not be the only possible mechanism by which such modifications confer greater efficiency of conversion. For example, as disclosed herein, certain modifications to oligonucleotides confer a greater improvement to the efficiency of conversion than other modifications.

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Efficiency of conversion is defined herein as the percentage of recovered substrate molecules that have undergone a conversion event. Depending on the nature of the target genetic material, e.g. the genome of a cell, efficiency could be represented as the proportion of cells or clones containing an extrachromosomal element that exhibit a particular phenotype. Alternatively, representative samples of the target genetic material can be sequenced to determine the percentage that have acquired the desire change. The oligonucleotides of the invention in different embodiments can alter DNA two, three, four, five, six, seven, eight, nine, ten, twelve, fifteen, twenty, thirty, and fifty or more fold more than control oligonucleotides. Such control oligonucleotides are oligonucleotides with fully phosphorothiolated linkages, oligonucleotides that are fully substituted with 2'-O-methyl analogs, a perfectly matched oligonucleotide that is fully complementary to a target sequence or a chimeric DNA-RNA double hairpin oligonucleotide such as disclosed in US Patent 5,565,350.

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In addition, for a given oligonucleotide length, additional modifications interfere with the ability of the oligonucleotide to act in concert with the cellular recombination or repair enzyme machinery

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which is necessary and required to mediate a targeted substitution, addition or deletion event in DNA. For example, fully phosphorothiolated or fully 2-O-methylated molecules are inefficient in targeted gene alteration.

The oligonucleotides of the invention as optimized for the purpose of targeted alteration of genetic material, including gene knockout or repair, are different in structure from antisense oligonucleotides that may possess a similar mixed chemical composition backbone. The oligonucleotides of the invention differ from such antisense oligonucleotides in chemical composition, structure, sequence, and in their ability to alter genomic DNA. Significantly, antisense oligonucleotides fail to direct targeted gene alteration. The oligonucleotides of the invention may target either strand of DNA and can include any component of the genome including, for example, intron and exon sequences. The preferred embodiment of the invention is a modified oligonucleotide that binds to the non-transcribed strand of a genomic DNA duplex. In other words, the preferred oligonucleotides of the invention target the sense strand of the DNA, i.e. the oligonucleotides of the invention are complementary to the non-transcribed strand of the target duplex DNA. The sequence of the non-transcribed strand of a DNA duplex is found in the mRNA produced from that duplex, given that mRNA uses uracil-containing nucleotides in place of thymine-containing nucleotides.

Moreover, the initial observation that single-stranded oligonucleotides comprising these modifications and lacking any particular triplex forming domain have reproducibly enhanced gene alteration activity in a variety of assay systems as compared to a chimeric RNA-DNA double-stranded hairpin control or single-stranded oligonucleotides comprising other backbone modifications was surprising. The single-stranded molecules of the invention totally lack the complementary RNA binding structure that stabilizes a normal chimeric double-stranded hairpin of the type disclosed in U.S. Patent 5,565,350 yet is more effective in producing targeted base conversion as compared to such a chimeric RNA-DNA double-stranded hairpin. In addition, the molecules of the invention lack any particular triplex forming domain involved in Hoogsteen interactions with the DNA double helix and required by other known oligonucleotides in other oligonucleotide-dependant gene conversion systems. Although the lack of these functional domains was expected to decrease the efficiency of an alteration in a sequence, just the opposite occurs: the efficiency of sequence alteration using the modified oligonucleotides of the invention is higher than the efficiency of sequence alteration using a chimeric RNA-DNA hairpin targeting the same sequence alteration. Moreover, the efficiency of sequence alteration or gene conversion directed by an unmodified oligonucleotide is many times lower as compared to a control chimeric RNA-DNA molecule or the modified oligonucleotides of the invention targeting the

same sequence alteration. Similarly, molecules containing at least 3 2'-O-methyl base analogs are about four to five fold less efficient as compared to an oligonucleotide having the same number of phosphorothioate linkages.

The oligonucleotides of the present invention for alteration of a single base are about 17 to about 121 nucleotides in length, preferably about 17 to about 74 nucleotides in length. Most preferably, however, the oligonucleotides of the present invention are at least about 25 bases in length, unless there are self-dimerization structures within the oligonucleotide. If the oligonucleotide has such an unfavorable structure, lengths longer than 35 bases are preferred. Oligonucleotides with modified ends both shorter and longer than certain of the exemplified, modified oligonucleotides herein function as gene repair or gene knockout agents and are within the scope of the present invention.

Once an oligomer is chosen, it can be tested for its tendency to self-dimerize, since self-dimerization may result in reduced efficiency of alteration of genetic information. Checking for self-dimerization tendency can be accomplished manually or, preferably, using a software program. One such program is Oligo Analyzer 2.0, available through Integrated DNA Technologies (Coralville, IA 52241) (http://www.idtdna.com); this program is available for use on the world wide web at

http://www.idtdna.com/program/oligoanalyzer/

oligoanalyzer.asp.

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For each oligonucleotide sequence input into the program, Oligo Analyzer 2.0 reports possible self-dimerized duplex forms, which are usually only partially duplexed, along with the free energy change associated with such self-dimerization. Delta G-values that are negative and large in magnitude, indicating strong self-dimerization potential, are automatically flagged by the software as "bad". Another software program that analyzes oligomers for pair dimer formation is Primer Select from DNASTAR, Inc., 1228 S. Park St., Madison, WI 53715, Phone: (608) 258-7420 (http://www.dnastar.com/products/PrimerSelect.html).

If the sequence is subject to significant self-dimerization, the addition of further sequence flanking the "repair" nucleotide can improve gene correction frequency.

Generally, the oligonucleotides of the present invention are identical in sequence to one strand of the target DNA, which can be either strand of the target DNA, with the exception of one or more targeted bases positioned within the DNA domain of the oligonucleotide, and preferably toward the middle between the modified terminal regions. Preferably, the difference in sequence of the oligonucleotide as compared to the targeted genomic DNA is located at about the middle of the oligonucleotide sequence. In a preferred embodiment, the oligonucleotides of the invention are complementary to the non-transcribed

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strand of a duplex. In other words, the preferred oligonucleotides target the sense strand of the DNA, i.e. the oligonucleotides of the invention are preferably complementary to the strand of the target DNA the sequence of which is found in the mRNA.

The oligonucleotides of the invention can include more than a single base change. In an oligonucleotide that is about a 70-mer, with at least one modified residue incorporated on the ends, as disclosed herein, multiple bases can be simultaneously targeted for change. The target bases may be up to 27 nucleotides apart and may not be changed together in all resultant plasmids in all cases. There is a frequency distribution such that the closer the target bases are to each other in the central DNA domain within the oligonucleotides of the invention, the higher the frequency of change in a given cell. Target bases only two nucleotides apart are changed together in every case that has been analyzed. The farther apart the two target bases are, the less frequent the simultaneous change. Thus, oligonucleotides of the invention may be used to repair or alter multiple bases rather than just one single base. For example, in a 74-mer oligonucleotide having a central base targeted for change, a base change event up to about 27 nucleotides away can also be effected. The positions of the altering bases within the oligonucleotide can be optimized using any one of the assays described herein. Preferably, the altering bases are at least about 8 nucleotides from one end of the oligonucleotide.

The oligonucleotides of the present invention can be introduced into cells by any suitable means. According to certain preferred embodiments, the modified oligonucleotides may be used alone. Suitable means, however, include the use of polycations, cationic lipids, liposomes, polyethylenimine (PEI), electroporation, biolistics, microinjection and other methods known in the art to facilitate cellular uptake. For plant cells, biolistic or particle bombardment methods are typically used. According to certain preferred embodiments of the present invention, isolated plant cells are treated in culture according to the methods of the invention, to mutate or repair a target gene. Alternatively, plant target DNA may be modified *in vitro* or in another cell type, including for example, yeast or bacterial cells and then introduced into a plant cell as, for example, a T-DNA. Plant cells thus modified may be used to regenerate the whole organism as, for example, in a plant having a desired targeted genomic change. In other instances, targeted genomic alteration, including repair or mutagenesis, may take place *in vivo* following direct administration of the modified, single-stranded oligonucleotides of the invention to a subject.

The single-stranded, modified oligonucleotides of the present invention have numerous applications as gene repair, gene modification, or gene knockout agents. Such oligonucleotides may be advantageously used, for example, to introduce or correct multiple point mutations. Each mutation leads to the addition, deletion or substitution of at least one base pair. The methods of the present invention

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offer distinct advantages over other methods of altering the genetic makeup of an organism, in that only the individually targeted bases are altered. No additional foreign DNA sequences are added to the genetic complement of the organism. Such agents may, for example, be used to develop plants with improved traits by rationally changing the sequence of selected genes in isolated cells and using these modified cells to regenerate whole plants having the altered gene. See, e.g., U.S. Patent 6,046,380 and U.S. Patent 5,905,185 incorporated herein by reference. Such plants produced using the compositions of the invention lack additional undesirable selectable markers or other foreign DNA sequences. Targeted base pair substitution or frameshift mutations introduced by an oligonucleotide in the presence of a cell-free extract also provides a way to modify the sequence of extrachromosomal elements, including, for example, plasmids, cosmids and artificial chromosomes. The oligonucleotides of the invention also simplify the production of plants having particular modified or inactivated genes. Altered plant model systems such as those produced using the methods and oligonucleotides of the invention are invaluable in determining the function of a gene and in evaluating drugs. The oligonucleotides and methods of the present invention may also be used to introduce molecular markers, including, for example, SNPs, RFLPs, AFLPs and CAPs.

The purified oligonucleotide compositions may be formulated in accordance with routine procedures depending on the target. For example, purified oligonucleotide can be used directly in a standard reaction mixture to introduce alterations into targeted DNA *in vitro* or where cells are the target as a composition adapted for bathing cells in culture or for microinjection into cells in culture. The purified oligonucleotide compositions may also be provided on coated microbeads for biolistic delivery into plant cells. Where necessary, the composition may also include a solubilizing agent. Generally, the ingredients will be supplied either separately or mixed together in single-use form, for example, as a dry, lyophilized powder or water-free concentrate. In general, dosage required for efficient targeted gene alteration will range from about 0.001 to 50,000 µg/kg target tissue, preferably between 1 to 250 µg/kg, and most preferably at a concentration of between 30 and 60 micromolar.

For cell administration, direct injection into the nucleus, biolistic bombardment, electroporation, liposome transfer and calcium phosphate precipitation may be used. In yeast, lithium acetate or spheroplast transformation may also be used. In a preferred method, the administration is performed with a liposomal transfer compound, e.g., DOTAP (Boehringer-Mannheim) or an equivalent such as lipofectin. The amount of the oligonucleotide used is about 500 nanograms in 3 micrograms of DOTAP per 100,000 cells. For electroporation, between 20 and 2000 nanograms of oligonucleotide per million cells to be electroporated is an appropriate range of dosages which can be increased to improve

efficiency of genetic alteration upon review of the appropriate sequence according to the methods described herein. For biolistic delivery, microbeads are generally coated with resuspended oligonucleotides, which range of oligonucleotide to microbead concentration can be similarly adjusted to improve efficiency as determined using one of the assay methods described herein, starting with about 0.05 to 1 microgram of oligonucleotide to 25 microgram of 1.0 micrometer gold beads or similar microcarrier.

Another aspect of the invention is a kit comprising at least one oligonucleotide of the invention. The kit may comprise an additional reagent or article of manufacture. The additional reagent or article of manufacture may comprise a delivery mechanism, cell extract, a cell, or a plasmid, such as one of those disclosed in the Figures herein, for use in an assay of the invention. Alternatively, the invention includes a kit comprising an isogenic set of cells in which each cell in the kit comprises a different altered amino acid for a target protein encoded by a targeted altered gene within the cell produced according to the methods of the invention.

Brief Description Of The Drawings

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Figure 1. Flow diagram for the generation of modified single-stranded oligonucleotides. The upper strands of chimeric oligonucleotides I and II are separated into pathways resulting in the generation of single-stranded oligonucleotides that contain (A) 2'-O-methyl RNA nucleotides or (B) phosphorothioate linkages. Fold changes in repair activity for correction of kan^s in the HUH7 cell-free extract are presented in parenthesis. HUH7 cells are described in Nakabayashi et al., Cancer Research 42: 3858-3863 (1982). Each single-stranded oligonucleotide is 25 bases in length and contains a G residue mismatched to the complementary sequence of the kan^s gene. The numbers 3, 6, 8, 10, 12 and 12.5 respectively indicate how many phosphorothioate linkages (S) or 2'-O-methyl RNA nucleotides (R) are at each end of the molecule. Hence oligo 12S/25G contains an all phosphorothioate backbone, displayed as a dotted line. Smooth lines indicate DNA residues, wavy lines indicate 2'-O-methyl RNA residues and the carat indicates the mismatched base site (G). Figure 1(C) provides a schematic plasmid indicating the sequence of the kan chimeric double-stranded hairpin oligonucleotide (left) and the sequence the tet chimeric double-stranded hairpin oligonucleotide used in other experiments. Figure 1(D) provides a flow chart of a kan experiment in which a chimeric double-stranded hairpin oligonucleotide is used.

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Figure 2. Genetic readout system for correction of a point mutation in plasmid pK^sm4021.

A mutant kanamycin gene harbored in plasmid pK^sm4021 is the target for correction by oligonucleotides.

The mutant G is converted to a C by the action of the oligo. Corrected plasmids confer resistance to kanamycin in *E.coli* (DH10B) after electroporation leading to the genetic readout and colony counts.

Figure 3: Target plasmid and sequence correction of a frameshift mutation by chimeric and single-stranded oligonucleotides. (A) Plasmid pT°Δ208 contains a single base deletion mutation at position 208 rendering it unable to confer tet resistance. The target sequence presented below indicates the insertion of a T directed by the oligonucleotides to re-establish the resistant phenotype. (B) DNA sequence confirming base insertion directed by Tet 3S/25G; the yellow highlight indicates the position of frameshift repair.

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Figure 4. DNA sequences of representative kan^r colonies. Confirmation of sequence alteration directed by the indicated molecule is presented along with a table outlining codon distribution. Note that 10S/25G and 12S/25G elicit both mixed and unfaithful gene repair. The number of clones sequenced is listed in parentheses next to the designation for the single-stranded oligonucleotide. A plus (+) symbol indicates the codon identified while a figure after the (+) symbol indicates the number of colonies with a particular sequence. TAC/TAG indicates a mixed peak. Representative DNA sequences are presented below the table with yellow highlighting altered residues.

Figure 5. Gene correction in HeLa cells. Representative oligonucleotides of the invention are co-transfected with the pCMVneo(')FIAsH plasmid (shown in Figure 9) into HeLa cells. Ligand is diffused into cells after co-transfection of plasmid and oligonucleotides. Green fluorescence indicates gene correction of the mutation in the antibiotic resistance gene. Correction of the mutation results in the expression of a fusion protein that carries a marker ligand binding site and when the fusion protein binds the ligand, a green fluorescence is emitted. The ligand is produced by Aurora Biosciences and can readily diffuse into cells enabling a measurement of corrected protein function; the protein must bind the ligand directly to induce fluorescence. Hence cells bearing the corrected plasmid gene appear green while "uncorrected" cells remain colorless.

Figure 6. Z-series imaging of corrected cells. Serial cross-sections of the HeLa cell represented in Figure 5 are produced by Zeiss 510 LSM confocal microscope revealing that the fusion protein is contained within the cell.

Figure 7. Hygromycin-eGFP target plasmids. (A) Plasmid pAURHYG(ins)GFP contains a single base insertion mutation between nucleotides 136 and 137, at codon 46, of the Hygromycin B coding sequence (cds) which is transcribed from the constitutive ADH1 promoter. The target sequence presented below indicates the deletion of an A and the substitution of a C for a T directed by the oligonucleotides to re-establish the resistant phenotype. (B) Plasmid pAURHYG(rep)GFP contains a

base substitution mutation introducing a G at nucleotide 137, at codon 46, of the Hygromycin B coding sequence (cds). The target sequence presented below the diagram indicates the amino acid conservative replacement of G with C, restoring gene function.

Figure 8. Oligonucleotides for correction of hygromycin resistance gene. The sequence of the oligonucleotides used in experiments to assay correction of a hygromycin resistance gene are shown. DNA residues are shown in capital letters, RNA residues are shown in lowercase and nucleotides with a phosphorothioate backbone are capitalized and underlined.

Figure 9. *pAURNeo(-)FIAsH plasmid*. This figure describes the plasmid structure, target sequence, oligonucleotides, and the basis for detection of the gene alteration event by fluorescence.

Figure 10. pYESHyg(x)eGFP plasmid. This plasmid is a construct similar to the pAURHyg(x)eGFP construct shown in Figure 7, except the promoter is the inducible GAL1 promoter. This promoter is inducible with galactose, leaky in the presence of raffinose, and repressed in the presence of dextrose.

Figure 11. pBI-HygeGFP plasmid. This plasmid is a construct based on the plasmids pBI101, pBI 101.2, pBI101.3 or pBI 121 available from Clontech in which HygeGFP replaces the beta-glucuronidase gene of the Clontech plasmids. The different Clontech plasmids vary by a reading frame shift relative to the polylinker, or the presence of the Cauliflower mosaic virus promoter.

The following examples are provided by way of illustration only, and are not intended to limit the scope of the invention disclosed herein.

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EXAMPLE 1 Assay Method For Base Alteration And Preferred Oligonucleotide Selection

In this example, single-stranded and double-hairpin oligonucleotides with chimeric backbones (see Figure 1 for structures (A and B) and sequences (C and D) of assay oligonucleotides) are used to correct a point mutation in the kanamycin gene of pK^sm4021 (Figure 2) or the tetracycline gene of $pT^s\Delta208$ (Figure 3). All kan oligonucleotides share the same 25 base sequence surrounding the target base identified for change, just as all tet oligonucleotides do. The sequence is given in Figures 1C and Figure 1D. Each plasmid contains a functional ampicillin gene. Kanamycin gene function is restored when a G at position 4021 is converted to a C (via a substitution mutation); tetracycline gene function is restored when a deletion at position 208 is replaced by a C (via frameshift mutation). A separate plasmid, pAURNeo(-)FIAsH (Figure 9), bearing the kan^s gene is used in the cell culture experiments. This plasmid was constructed by inserting a synthetic expression cassette containing a neomycin phosphotransferase

(kanamycin resistance) gene and an extended reading frame that encodes a receptor for the FIAsH ligand into the pAUR123 shuttle vector (Panvera Corp., Madison, WI). The resulting construct replicates in *S. cerevisiae* at low copy number, confers resistance to aureobasidinA and constitutively expresses either the Neo+/FIAsH fusion product (after alteration) or the truncated Neo-/FIAsH product (before alteration) from the ADH1 promoter. By extending the reading frame of this gene to code for a unique peptide sequence capable of binding a small ligand to form a fluorescent complex, restoration of expression by correction of the stop codon can be detected in real time using confocal microscopy.

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Additional constructs can be made to test additional gene alteration events or for specific use in different expression systems. For example, alternative comparable plant plasmids or integration vectors such as, e.g. those based on T-DNA, can be constructed for stable expression in plant cells according to the disclosures herein. Such constructs would use a plant specific promoter such as, e.g., cauliflower mosaic virus 35S promoter, to replace the promoters directing expression of the neo, hyg or aureobasidinA resistance gene disclosed herein, including for example, in Figures 7B, 9 and 10 herein. Moreover, the green fluorescent protein (GFP) sequence used herein may be modified to increase expression in plant cells such as Arabidopsis and the other plants disclosed herein as described in Haseloff et al., Proc. Natl.Acad. Sci. 94(6): 2122-7 (1997), Rouwendal et al. Plant Mol. Biol. 33(6): 989-99 (1997) and Hu et al. FEBS Lett. 369(2-3): 331-4 (1995). Codon usage for optimal expression of GFP in plants results from increasing the frequency of codons with a C or a G in the third position from 32 to about 60%. Specific constructs are disclosed and can be used as follows with such plant specific alterations.

We also construct three mammalian expression vectors, pHyg(rep)eGFP, pHyg(Δ)eGFP, pHyg(ins)eGFP, that contain a substitution mutation at nucleotide 137 of the hygromycin-B coding sequence. (rep) indicates a T137→G replacement, (Δ) represents a deletion of the G137 and (ins) represents an A insertion between nucleotides 136 and 137. All point mutations create a nonsense termination codon at residue 46. We use pHYGeGFP plasmid (Invitrogen, CA) DNA as a template to introduce the mutations into the hygromycin-eGFP fusion gene by a two step site-directed mutagenesis PCR protocol. First, we generate overlapping 5' and a 3' amplicons surrounding the mutation site by PCR for each of the point mutation sites. A 215 bp 5' amplicon for the (rep), (Δ) or (ins) was generated by polymerization from oligonucleotide primer HygEGFPf (5'-AATACGACTCACTATAGG-3') to primer Hygrepr (5'GACCTATCCACGCCCTCC-3'), HygΔr (5'-GACTATCCACGCCCTCC-3'), or Hyginsr (5'-GACATTATCCACGCCCTCC-3'), respectively. We generate a 300bp 3' amplicon for the (rep), (Δ) or (ins) by polymerization from oligonucleotide primers Hygrepf (5'-CTGGGATAGGTCCTGCGG-3'), HygΔf

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(5'-CGTGGATAGTCCTGCGG-3'), Hyginsf (5'-CGTGGATAATGTCCTGCGG-3'), respectively to primer HygEGFPr (5'-AAATCACGCCATGTAGTG-3'). We mix 20 ng of each of the resultant 5' and 3' overlapping amplicon mutation sets and use the mixture as a template to amplify a 523 bp fragment of the Hygromycin gene spanning the KpnI and RsrII restriction endonuclease sites. We use the Expand PCR system (Roche) to generate all amplicons with 25 cycles of denaturing at 94°C for 10 seconds, annealing at 55°C for 20 seconds and elongation at 68°C for 1 minute. We digest 10 µg of vector pHYGeGFP and 5 µg of the resulting fragments for each mutation with KpnI and RsrII (NEB) and gel purify the fragment for enzymatic ligation. We ligate each mutated insert into pHYGeGFP vector at 3:1 molar ratio using T4 DNA ligase (Roche). We screen clones by restriction digest, confirm the mutation by Sanger dideoxy chain termination sequencing and purify the plasmid using a Qiagen maxiprep kit.

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Oligonucleotide synthesis and cells. Chimeric oligonucleotides and single-stranded oligonucleotides (including those with the indicated modifications) are synthesized using available phosphoramidites on controlled pore glass supports. After deprotection and detachment from the solid support, each oligonucleotide is gel-purified using, for example, procedures such as those described in Gamper et al., Biochem. 39, 5808-5816 (2000) and the concentrations determined spectrophotometrically (33 or 40 µg/ml per A₂₆₀ unit of single-stranded or hairpin oligomer). HUH7 cells are grown in DMEM, 10% FBS, 2mM glutamine, 0.5% pen/strep. The *E.coli* strain, DH10B, is obtained from Life Technologies (Gaithersburg, MD); DH10B cells contain a mutation in the RECA gene (*recA*).

Cell-free extracts. Although this portion of this example is directed to mammalian systems, similar extracts from plants can be prepared as disclosed elsewhere in this application and used as disclosed in this example. We prepare cell-free extracts from HUH7 cells or other mammalian cells, as follows. We employ this protocol with essentially any mammalian cell including, for example, H1299 cells (human epithelial carcinoma, non-small cell lung cancer), C127I (immortal murine mammary epithelial cells), MEF (mouse embryonic fibroblasts), HEC-1-A (human uterine carcinoma), HCT15 (human colon cancer), HCT116 (human colon carcinoma), LoVo (human colon adenocarcinoma), and HeLa (human cervical carcinoma). We harvest approximately 2x10⁸ cells. We then wash the cells immediately in cold hypotonic buffer (20 mM HEPES, pH7.5; 5 mM KCl; 1.5 mM MgCl₂; 1 mM DTT) with 250 mM sucrose. We then resuspend the cells in cold hypotonic buffer without sucrose and after 15 minutes we lyse the cells with 25 strokes of a Dounce homogenizer using a tight fitting pestle. We incubate the lysed cells for 60 minutes on ice and centrifuge the sample for 15 minutes at 12000xg. The cytoplasmic fraction is enriched with nuclear proteins due to the extended co-incubation of the fractions following cell breakage.

We then immediately aliquote and freeze the supernatant at -80°C. We determine the protein concentration in the extract by the Bradford assay.

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We also perform these experiments with cell-free extracts obtained from fungal cells, including, for example, *S. cerevisiae* (yeast), *Ustilago maydis*, and *Candida albicans*. For example, we grow yeast cells into log phase in 2L YPD medium for 3 days at 30°C. We then centrifuge the cultures at 5000xg, resuspend the pellets in a 10% sucrose, 50 mM Tris, 1mM EDTA lysis solution and freeze them on dry ice. After thawing, we add KCl, spermidine and lyticase to final concentrations of 0.25 mM, 5 mM and 0.1 mg/ml, respectively. We incubate the suspension on ice for 60 minutes, add PMSF and Triton X100 to final concentrations of 0.1 mM and 0.1% and continue to incubate on ice for 20 minutes. We centrifuge the lysate at 3000xg for 10 minutes to remove larger debris. We then remove the supernatant and clarify it by centrifuging at 30000xg for 15 minutes. We then add glycerol to the clarified extract to a concentration of 10% (v/v) and freeze aliquots at -80°C. We determine the protein concentration of the extract by the Bradford assay.

Reaction mixtures of 50 µl are used, consisting of 10-30 µg protein of cell-free extract, which can be optionally substituted with purified proteins or enriched fractions, about 1.5 µg chimeric double-hairpin oligonucleotide or 0.55 µg single-stranded molecule (3S/25G or 6S/25G, see Figure 1), and 1 µg of plasmid DNA (see Figures 2 and 3) in a reaction buffer of 20 mM Tris, pH 7.4, 15 mM MgCl₂, 0.4 mM DTT, and 1.0 mM ATP. Reactions are initiated with extract and incubated at 30°C for 45 min. The reaction is stopped by placing the tubes on ice and then immediately deproteinized by two phenol/chloroform (1:1) extractions. Samples are then ethanol precipitated. The nucleic acid is pelleted at 15,000 r.p.m. at 4°C for 30 min., is washed with 70% ethanol, resuspended in 50 μ l H₂0, and is stored at -20°C. 5 µl of plasmid from the resuspension (~100 ng) was transfected in 20 µl of DH10B cells by electroporation (400 V, 300 μ F, 4 k Ω) in a Cell-Porator apparatus (Life Technologies). After electroporation, cells are transferred to a 14 ml Falcon snap-cap tube with 2 ml SOC and shaken at 37°C for 1 h. Enhancement of final kan colony counts is achieved by then adding 3 ml SOC with 10 µg/ml kanamycin and the cell suspension is shaken for a further 2 h at 37°C. Cells are then spun down at 3750 x g and the pellet is resuspended in 500 µI SOC. 200 µI is added undiluted to each of two kanamycin (50 µg/ml) agar plates and 200 µl of a 10⁵ dilution is added to an ampicillin (100 µg/ml) plate. After overnight 37°C incubation, bacterial colonies are counted using an Accucount 1000 (Biologics). Gene conversion effectiveness is measured as the ratio of the average of the kan colonies on both plates per amp colonies multiplied by 10⁻⁵ to correct for the amp dilution.

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The following procedure can also be used. 5 μ I of resuspended reaction mixtures (total volume 50 μ I) are used to transform 20 μ I aliquots of electro-competent DH10B bacteria using a Cell-Porator apparatus (Life Technologies). The mixtures are allowed to recover in 1 ml SOC at 37°C for 1 hour at which time 50 μ g/ml kanamycin or 12 μ g/ml tetracycline is added for an additional 3 hours. Prior to plating, the bacteria are pelleted and resuspended in 200 μ I of SOC. 100 μ I aliquots are plated onto kan or tet agar plates and 100 μ I of a 10⁻⁴ dilution of the cultures are concurrently plated on agar plates containing 100 μ g/ml of ampicillin. Plating is performed in triplicate using sterile Pyrex beads. Colony counts are determined by an Accu-count 1000 plate reader (Biologics). Each plate contains 200-500 ampicillin resistant colonies or 0-500 tetracycline or kanamycin resistant colonies. Resistant colonies are selected for plasmid extraction and DNA sequencing using an ABI Prism kit on an ABI 310 capillary sequencer (PE Biosystems).

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Chimeric single-stranded oligonucleotides. In Figure 1 the upper strands of chimeric oligonucleotides I and II are separated into pathways resulting in the generation of single-stranded oligonucleotides that contain (Figure 1A) 2'-O-methyl RNA nucleotides or (Figure 1B) phosphorothioate linkages. Fold changes in repair activity for correction of kan^s in the HUH7 cell-free extract are presented in parenthesis. Each single-stranded oligonucleotide is 25 bases in length and contains a G residue mismatched to the complementary sequence of the kan^s gene.

Molecules bearing 3, 6, 8, 10 and 12 phosphorothioate linkages in the terminal regions at each end of a backbone with a total of 24 linkages (25 bases) are tested in the kan^s system. Alternatively, molecules bearing 2, 4, 5, 7, 9 and 11 in the terminal regions at each end are tested. The results of one such experiment, presented in Table 1 and Figure 1B, illustrate an enhancement of correction activity directed by some of these modified structures. In this illustrative example, the most efficient molecules contained 3 or 6 phosphorothioate linkages at each end of the 25-mer; the activities are approximately equal (molecules IX and X with results of 3.09 and 3.7 respectively). A reduction in alteration activity may be observed as the number of modified linkages in the molecule is further increased. Interestingly, a single-strand molecule containing 24 phosphorothioate linkages is minimally active suggesting that this backbone modification when used throughout the molecule supports only a low level of targeted gene repair or alteration. Such a non-altering, completely modified molecule can provide a baseline control for determining efficiency of correction for a specific oligonucleotide molecule of known sequence in defining the optimum oligonucleotide for a particular alteration event.

The efficiency of gene repair directed by phosphorothioate-modified, single-stranded molecules, in a length dependent fashion, led us to examine the length of the RNA modification used in

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the original chimera as it relates to correction. Construct III represents the "RNA-containing" strand of chimera I and, as shown in Table 1 and Figure 2A, it promotes inefficient gene repair. But, as shown in the same figure, reducing the RNA residues on each end from 10 to 3 increases the frequency of repair. At equal levels of modification, however, 25-mers with 2'-O-methyl ribonucleotides were less effective gene repair agents than the same oligomers with phosphorothioate linkages. These results reinforce the fact that an RNA containing oligonucleotide is not as effective in promoting gene repair or alteration as a modified DNA oligonucleotide.

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Repair of the kanamycin mutation requires a G→C exchange. To confirm that the specific desired correction alteration was obtained, colonies selected at random from multiple experiments are processed and the isolated plasmid DNA is sequenced. As seen in Figure 4, colonies generated through the action of the single-stranded molecules 3S/25G (IX), 6S/25G (X) and 8S/25G (XI) respectively contained plasmid molecules harboring the targeted base correction. While a few colonies appeared on plates derived from reaction mixtures containing 25-mers with 10 or 12 thioate linkages on both ends, the sequences of the plasmid molecules from these colonies contain nonspecific base changes. In these illustrative examples, the second base of the codon is changed (see Figure 3). These results show that modified single-strands can direct gene repair, but that efficiency and specificity are reduced when the 25-mers contain 10 or more phosphorothioate linkages at each end.

In Figure 1, the numbers 3, 6, 8, 10, 12 and 12.5 respectively indicate how many phosphorothioate linkages (S) or 2'-O-methyl RNA nucleotides (R) are at each end of the examplified molecule although other molecules with 2, 4, 5, 7, 9 and 11 modifications at each end can also be tested. Hence oligo 12S/25G represents a 25-mer oligonucleotide which contains 12 phosphorothioate linkages on each side of the central G target mismatch base producing a fully phosphorothioate linked backbone, displayed as a dotted line. The dots are merely representative of a linkage in the figure and do not depict the actual number of linkages of the oligonucleotide. Smooth lines indicate DNA residues, wavy lines indicate 2'-O-methyl RNA residues and the carat indicates the mismatched base site (G).

Correction of a mutant kanamycin gene in cultured mammalian cells. Although this portion of this example is directed to cultured mammalian cells, comparable methods may be used using cultured plant cells or protoplasts of those cells from the plant species disclosed herein. The experiments are performed using different eukaryotic cells including plant and mammalian cells, including, for example, 293 cells (transformed human primary kidney cells), HeLa cells (human cervical carcinoma), and H1299 (human epithelial carcinoma, non-small cell lung cancer). HeLa cells are grown at 37°C and 5% CO₂ in a humidified incubator to a density of 2 x 10⁵ cells/ml in an 8 chamber slide (Lab-Tek). After replacing the

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regular DMEM with Optimem, the cells are co-transfected with 10 µg of plasmid pAURNeo(-)FIAsH and 5 µg of modified single-stranded oligonucleotide (3S/25G) that is previously complexed with 10 µg lipofectamine, according to the manufacturer's directions (Life Technologies). The cells are treated with the liposome-DNA-oligo mix for 6 hrs at 37°C. Treated cells are washed with PBS and fresh DMEM is added. After a 16-18 hr recovery period, the culture is assayed for gene repair. The same oligonucleotide used in the cell-free extract experiments is used to target transfected plasmid bearing the kan^s gene. Correction of the point mutation in this gene eliminates a stop codon and restores full expression. This expression can be detected by adding a small non-fluorescent ligand that bound to a C-C-R-E-C-C sequence in the genetically modified carboxy terminus of the kan protein, to produce a highly fluorescent complex (FIAsH system, Aurora Biosciences Corporation). Following a 60 min incubation at room temperature with the ligand (FIAsH-EDT2), cells expressing full length kan product acquire an intense green fluorescence detectable by fluorescence microscopy using a fluorescein filter set. Similar experiments are performed using the HygeGFP target as described in Example 2 with a variety of mammalian cells, including, for example, COS-1 and COS-7 cells (African green monkey), and CHO-K1 cells (Chinese hamster ovary). The experiments are also performed with PG12 cells (rat pheochromocytoma) and ES cells (human embryonic stem cells).

Summary of experimental results. Tables 1, 2 and 3 respectively provide data on the efficiency of gene repair directed by single-stranded oligonucleotides. Table 1 presents data using a cell-free extract from human liver cells (HUH7) to catalyze repair of the point mutation in plasmid pkan^sm4021 (see Figure 1). Table 2 illustrates that the oligomers are not dependent on MSH2 or MSH3 for optimal gene repair activity. Table 3 illustrates data from the repair of a frameshift mutation (Figure 3) in the tet gene contained in plasmid pTetΔ208. Table 4 illustrates data from repair of the pkan^sm4021 point mutation catalyzed by plant cell extracts prepared from canola and musa (banana). Colony numbers are presented as kan^r or tet^r and fold increases (single strand versus double hairpin) are presented for kan^r in Table 1.

Figure 5A is a confocal picture of HeLa cells expressing the corrected fusion protein from an episomal target. Gene repair is accomplished by the action of a modified single-stranded oligonucleotide containing 3 phosphorothicate linkages at each end (3S/25G). Figure 5B represents a "Z-series" of HeLa cells bearing the corrected fusion gene. This series sections the cells from bottom to top and illustrates that the fluorescent signal is "inside the cells".

Results. In summary, we have designed a novel class of single-stranded oligonucleotides with backbone modifications at the termini and demonstrate gene repair/conversion

activity in mammalian and plant cell-free extracts. We confirm that the all DNA strand of the RNA-DNA double-stranded double hairpin chimera is the active component in the process of gene repair. In some cases, the relative frequency of repair by the novel oligonucleotides of the invention is elevated approximately 3-4-fold in certain embodiments when compared to frequencies directed by chimeric RNA-DNA double hairpin oligonucleotides.

This strategy centers around the use of extracts from various sources to correct a mutation in a plasmid using a modified single-stranded or a chimeric RNA-DNA double hairpin oligonucleotide. A mutation is placed inside the coding region of a gene conferring antibiotic resistance in bacteria, here kanamycin or tetracycline. The appearance of resistance is measured by genetic readout in *E.coli* grown in the presence of the specified antibiotic. The importance of this system is that both phenotypic alteration and genetic inheritance can be measured. Plasmid pKsm4021 contains a mutation (T→G) at residue 4021 rendering it unable to confer antibiotic resistance in *E.coli*. This point mutation is targeted for repair by oligonucleotides designed to restore kanamycin resistance. To avoid concerns of plasmid contamination skewing the colony counts, the directed correction is from G→C rather than G→T (wild-type). After isolation, the plasmid is electroporated into the DH10B strain of *E.coli*, which contains inactive RecA protein. The number of kanamycin colonies is counted and normalized by ascertaining the number of ampicillin colonies, a process that controls for the influence of electroporation. The number of colonies generated from three to five independent reactions was averaged and is presented for each experiment. A fold increase number is recorded to aid in comparison.

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The original RNA-DNA double hairpin chimera design, e.g., as disclosed in U.S. Patent 5,565,350, consists of two hybridized regions of a single-stranded oligonucleotide folded into a double hairpin configuration. The double-stranded targeting region is made up of a 5 base pair DNA/DNA segment bracketed by 10 base pair RNA/DNA segments. The central base pair is mismatched to the corresponding base pair in the target gene. When a molecule of this design is used to correct the kans mutation, gene repair is observed (I in Figure 1A). Chimera II (Figure 1B) differs partly from chimera I in that only the DNA strand of the double hairpin is mismatched to the target sequence. When this chimera was used to correct the kans mutation, it was twice as active. In the same study, repair function could be further increased by making the targeting region of the chimera a continuous RNA/DNA hybrid.

Frame shift mutations are repaired. By using plasmid pTs \(\triangle 208\), described in Figure 1(C) and Figure 3, the capacity of the modified single-stranded molecules that showed activity in correcting a point mutation, can be tested for repair of a frameshift. To determine efficiency of correction of the mutation, a chimeric oligonucleotide (Tet I), which is designed to insert a T residue at position 208, is

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used. A modified single-stranded oligonucleotide (Tet IX) directs the insertion of a T residue at this same site. Figure 3 illustrates the plasmid and target bases designated for change in the experiments. When all reaction components are present (extract, plasmid, oligomer), tetracycline resistant colonies appear. The colony count increases with the amount of oligonucleotide used up to a point beyond which the count falls off (Table 3). No colonies above background are observed in the absence of either extract or oligonucleotide, nor when a modified single-stranded molecule bearing perfect complementarity is used. Figure 3 represents the sequence surrounding the target site and shows that a T residue is inserted at the correct site. We have isolated plasmids from fifteen colonies obtained in three independent experiments and each analyzed sequence revealed the same precise nucleotide insertion. These data suggest that the single-stranded molecules used initially for point mutation correction can also repair nucleotide deletions.

Comparison of phosphorothioate oligonucleotides to 2'-O-methyl substituted oligonucleotides. From a comparison of molecules VII and XI, it is apparent that gene repair is more subject to inhibition by RNA residues than by phosphorothioate linkages. Thus, even though both of these oligonucleotides contain an equal number of modifications to impart nuclease resistance, XI (with 16 phosphorothioate linkages) has good gene repair activity while VII (with 16 2'-O-methyl RNA residues) is inactive. Hence, the original chimeric double hairpin oligonucleotide enabled correction directed, in large part, by the strand containing a large region of contiguous DNA residues.

Oligonucleotides can target multiple nucleotide alterations within the same template. The ability of individual single-stranded oligonucleotides to correct multiple mutations in a single target template is tested using the plasmid pKsm4021 and the following single-stranded oligonucleotides modified with 3 phosphorothioate linkages at each end (indicated as underlined nucleotides): Oligo1 is a 25-mer with the sequence TTCGATAAGCCTATGCTGACCCGTG corrects the original mutation present in the kanamycin resistance gene of pKsm4021 as well as directing another alteration 2 basepairs away in the target sequence (both indicated in boldface); Oligo2 is a 70-mer with the 5'-end sequence TTCGGCTACGACTGGGCACAACAGACAATTGGC with the remaining nucleotides being completely complementary to the kanamycin resistance gene and also ending in 3 phosphorothioate linkages at the 3' end. Oligo2 directs correction of the mutation in pKsm4021 as well as directing another alteration 21 basepairs away in the target sequence (both indicated in boldface).

We also use additional oligonucleotides to assay the ability of individual oligonucleotides to correct multiple mutations in the pKsM4021 plasmid. These include, for example, a second 25-mer that alters two nucleotides that are three nucleotides apart with the sequence 5'-

TTGTGCCCAGTCGTATCCGAATAGC-3'; a 70-mer that alters two nucleotides that are 21 nucleotides apart with the sequence 5'-CATCAGAGCAGCCAATTGTCTGTTGTGCCCAGTCGTAGCCGAA
TAGCCTCTCCACCCAAGCGGCCGGAGA-3'; and another 70-mer that alters two nucleotides that are 21 nucleotides apart with the sequence 5'-

GCTGACAGCCGGAACACGGCGGCATCAGAGCAGCCAATTGTCTGTTGTGCCCAGTCGTAGCCGAAT AGCCT-3'. The nucleotides in the oligonucleotides that direct alteration of the target sequence are underlined and in boldface. These oligonucleotides are modified in the same way as the other oligonucleotides of the invention.

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We assay correction of the original mutation in pKsm4021 by monitoring kanamycin resistance (the second alterations which are directed by Oligo2 and Oligo3 are silent with respect to the kanamycin resistance phenotype). In addition, in experiments with Oligo2, we also monitor cleavage of the resulting plasmids using the restriction enzyme Tsp509l which cuts at a specific site present only when the second alteration has occurred (at ATT in Oligo2). We then sequence these clones to determine whether the additional, silent alteration has also been introduced. The results of an analysis are presented below:

Oligo1 (25-mer)	Oligo2 (70-mer)	
9	7	
0	2	
4	1	
	Oligo1 (25-mer) 9 0 4	

Nuclease sensitivity of unmodified DNA oligonucleotide. Electrophoretic analysis of nucleic acid recovered from the cell-free extract reactions conducted here confirm that the unmodified single-stranded 25-mer did not survive incubation whereas greater than 90% of the terminally modified oligos did survive (as judged by photo-image analyses of agarose gels).

Plant extracts direct repair. The modified single-stranded constructs can be tested in plant cell extracts. We have observed gene alteration using extracts from multiple plant sources, including, for example, Arabidopsis, tobacco, banana, maize, soybean, canola, wheat, spinach as well as spinach chloroplast extract or extracts made from other plant cells disclosed herein. We prepare the extracts by grinding plant tissue or cultured cells under liquid nitrogen with a mortar and pestle. We extract 3 ml of the ground plant tissue with 1.5 ml of extraction buffer (20 mM HEPES, pH7.5; 5 mM KCl;

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1.5 mM MgCl₂; 10 mM DTT; and 10% [v/v] glycerol). Some plant cell-free extracts also include about 1% (w/v) PVP. We then homogenize the samples with 15 strokes of a Dounce homogenizer. Following homogenization, we incubate the samples on ice for 1 hour and centrifuge at 3000 x g for 5 minutes to remove plant cell debris. We then determine the protein concentration in the supernatants (extracts) by Bradford assay. We dispense 100 µg (protein) aliquots of the extracts which we freeze in a dry ice-ethanol bath and store at -80°C.

We describe experiments using two sources here: a dicot (canola) and a monocot (banana, *Musa acuminata* cv. Rasthali). Each vector directs gene repair of the kanamycin mutation (Table 4); however, the level of correction is elevated 2-3 fold relative to the frequency observed with the chimeric oligonucleotide. These results are similar to those observed in the mammalian system wherein a significant improvement in gene repair occurred when modified single-stranded molecules were used.

Tables are attached hereto.

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Table I

Gene repair activity is directed by single-stranded oligonucleotides.

Oligonucleotide	Plasmid	Extract (ug)	kan ^r colonies	Fold increase
I	pK ^S m4021	10	300	
I	- 1	20	418	1.0x
Ц	İ	10	537	
П		20	748	1.78x
m		10	3	•
Ш	İ	20	5	0.01x
IV		10	112	
IV	į	20	96	0.22x
V		10	217	
V	ļ	20	342	0.81x
VI	ļ	10	6	
VI	.	20	39	0.093x
VII		10	0	
VII		20	0	0x
VIII		10	. 3	•
VIII	·	20	5	0.01x
IX		10	936	
ΙX		20	1295	3.09x
X		10	1140	
X		20	1588	3.7x
XI	l	10	480	
XI		20	681	1.6x
XII		10	18	
XII		20	25	0.059x
XШ		10	0	
XIII		20	4	0.009x
- *		20	0	•
I	▼	-	0 .	

Plasmid pK⁴m4021 (1µg), the indicated oligonucleotide (1.5 µg chimeric oligonucleotide or 0.55 µg single-stranded oligonucleotide; molar ratio of oligo to plasmid of 360 to 1) and either 10 or 20 µg of HUH7 cell-free extract were incubated 45 min at 37°C. Isolated plasmid DNA was electroporated into *E. coli* (strain DH10B) and the number of kan^r colonies counted. The data represent the number of kanamycin resistant colonies per 10⁶ ampicillin resistant colonies generated from the same reaction and is the average of three

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experiments (standard deviation usually less than +/- 15%). Fold increase is defined relative to 418 kan^r colonies (second reaction) and in all reactions was calculated using the 20µg sample.

Table II

Modified single-stranded oligomers are not dependent on MSH2 or MSH3 for optimal gene repair activity.

A. Oligonucleotide Pla	smid Extract	<u>kan^r colonies</u>
A. Oligonucieoude Fia	HUH7	637
IX (3S/25G)	HUH7	836
X (6S/25G)	MEF2 [→]	781
IX	MEF2 ^{-/-}	676
X	MEF3	582
IX	MEF3	530
X	MEF**	332
· IX	MEF*/*	497
X		10
-	MEF2*	5
-	MEF3	14
-	₩EF ^{*/*}	14 ,

Chimeric oligonucleotide (1.5 µg) or modified single-stranded oligonucleotide (0.55 µg) was incubated with 1µg of plasmid pK*m4021 and 20µg of the indicated extracts. MEF represents mouse embryonic fibroblasts with either MSH2 (2^{-/-}) or MSH3 (3^{-/-}) deleted.

MEF^{-/-} indicates wild-type mouse embryonic fibroblasts. The other reaction components were then added and processed through the bacterial readout system. The data represent the number of kanamycin resistant colonies per 10⁶ ampicillin resistant colonies.

Table III

Frameshift mutation repair is directed by single-stranded oligonucleotides

Oligonucleotide	Plasmid	Extract	tet ^r colonies
Tet IX (3S/25A; 0.5 μg)	pT ⁴ Δ208 (1μg)		- 0
-		20µg	0
Tet IX (0.5 μg)		1	48
Tet IX (1.5 μg)		1	130
Tet IX (2.0 μg)			68
Tet I (chimera; 1.5 μg)	▼	★	48

Each reaction mixture contained the indicated amounts of plasmid and oligonucleotide. The extract used for these experiments came from HUH7 cells. The data represent the number of tetracycline resistant colonies per 10⁶ ampicillin resistant colonies generated from the same reaction and is the average of 3 independent experiments. Tet I is a chimeric oligonucleotide and Tet IX is a modified single-stranded oligonucleotide that are designed to insert a T residue at position 208 of pT⁶Δ208. These oligonucleotides are equivalent to structures I and IX in Figure 2.

Table IV

Plant cell-free extracts support gene repair by single-stranded oligonucleotides

Oli-ampleotide	Plasmid	Extract	kan ^r colonies
Oligonucleotide	pK ^S m4021	30μg Canola	337
II (chimera)	pk. m4021	Canola	763
IX (3S/25G)		-	882
X (6S/25G)		Canola	203
П		Musa	
ΪX		Musa	343
X		Musa	746
Λ		Canola	0
•		Musa	0
-	,	- Canola	0
IX		- Musa	0
X	. ↓	- Musa	•

Canola or Musa cell-free extracts were tested for gene repair activity on the kanamycin-sensitive gene as previously described in (18). Chimeric oligonucleotide II (1.5 µg) and modified single-stranded oligonucleotides IX and X (0.55µg) were used to correct pK^Sm4021. Total number of kan^r colonies are present per 10⁷ ampicillin resistant colonies and represent an average of four independent experiments.

Table V

Gene repair activity in cell-free extracts prepared from yeast (Saccharomyces cerevisiae)

ell-type	Plasmid	Chimeric Oligo	SS Oliga	SS Oligo kan'/amp' x 106
ltype	pKan m4021	Ing	į	0.36
Wild type ARAD52		Bill	8 1 1	0.81 10.72
VD52		•	Jµg	17.41
ÆS1		1µg		2.02
4S1	~		3rl	3.23

In this experiment, the kan' gene in pKan' 4021 is corrected by either a chimeric double-hairpin oligonucleotide or a single-stranded oligonucleotide comaining three thioate linkages at each end (3S/25G).

EXAMPLE 2 Yeast Cell Targeting Assay Method for Base Alteration and Preferred Oligonucleotide Selection

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In this example, single-stranded oligonucleotides with modified backbones and double-hairpin oligonucleotides with chimeric, RNA-DNA backbones are used to measure gene repair using two episomal targets with a fusion between a hygromycin resistance gene and eGFP as a target for gene repair. These plasmids are pAURHYG(rep)GFP, which contains a point mutation in the hygromycin resistance gene (Figure 7), pAURHYG(ins)GFP, which contains a single-base insertion in the hygromycin resistance gene (Figure 7) and pAURHYG(Δ)GFP which has a single base deletion. We also use the plasmid containing a wild-type copy of the hygromycin-eGFP fusion gene, designated pAURHYG(wt)GFP, as a control. These plasmids also contain an aureobasidinA resistance gene. In pAURHYG(rep)GFP, hygromycin resistance gene function and green fluorescence from the eGFP protein are restored when a G at position 137, at codon 46 of the hygromycin B coding sequence, is converted to a C thus removing a premature stop codon in the hygromycin resistance gene coding region. In pAURHYG(ins)GFP, hygromycin resistance gene function and green fluorescence from the eGFP protein are restored when an A inserted between nucleotide positions 136 and 137, at codon 46 of the hygromycin B coding sequence, is deleted and a C is substituted for the T at position 137, thus correcting a frameshift mutation and restoring the reading frame of the hygromycin-eGFP fusion gene.

We synthesize the set of three yeast expression constructs pAURHYG(rep)eGFP, pAURHYG(Δ)eGFP, pAURHYG(ins)eGFP, that contain a point mutation at nucleotide 137 of the hygromycin-B coding sequence as follows. (rep) indicates a T137→G replacement, (Δ) represents a deletion of the G137 and (ins) represents an A insertion between nucleotides 136 and 137. We construct this set of plasmids by excising the respective expression cassettes by restriction digest from pHyg(x)EGFP and ligation into pAUR123 (Panvera, CA). We digest 10 μg pAUR123 vector DNA, as well as, 10 μg of each pHyg(x)EGFP construct with KpnI and Sall (NEB). We gel purify each of the DNA fragments and prepare them for enzymatic ligation. We ligate each mutated insert into pHygEGFP vector at 3:1 molar ratio using T4 DNA ligase (Roche). We screen clones by restriction digest, confirm by Sanger dideoxy chain termination sequencing and purify using a Qiagen maxiprep kit.

We use this system to assay the ability of five oligonucleotides (shown in Figure 8) to support correction under a variety of conditions. The oligonucleotides which direct correction of the mutation in pAURHYG(rep)GFP can also direct correction of the mutation in pAURHYG(ins)GFP. Three of the four oligonucleotides (HygE3T/25, HygE3T/74 and HygGG/Rev) share the same 25-base sequence surrounding the base targeted for alteration. HygGG/Rev is an RNA-DNA chimeric double hairpin

oligonucleotide of the type described in the prior art. One of these oligonucleotides, HygE3T/74, is a 74-base oligonucleotide with the 25-base sequence centrally positioned. The fourth oligonucleotide, designated HygE3T/74 α , is the reverse complement of HygE3T/74. The fifth oligonucleotide, designated Kan70T, is a non-specific, control oligonucleotide which is not complementary to the target sequence. Alternatively, an oligonucleotide of identical sequence but lacking a mismatch to the target or a completely thioate modified oligonucleotide or a completely 2-0-methylated modified oligonucleotide may be used as a control. Alternatively, oligonucleotides containing one, two, three, four, five, six, eight, ten or more LNA modifications on at least one of the two termini (and preferrably the 3' terminus) may be used in different embodiments.

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Oligonucleotide synthesis and cells. We synthesized and purified the chimeric, doublehairpin oligonucleotides and single-stranded oligonucleotides (including those with the indicated modifications) as described in Example 1. Plasmids used for assay were maintained stably in yeast (Saccharomyces cerevisiae) strain LSY678 MAT \alpha at low copy number under aureobasidin selection. Plasmids and oligonucleotides are introduced into yeast cells by electroporation as follows: to prepare electrocompetent yeast cells, we inoculate 10 ml of YPD media from a single colony and grow the cultures overnight with shaking at 300 rpm at 30°C. We then add 30 ml of fresh YPD media to the overnight cultures and continue shaking at 30°C until the OD_{son} was between 0.5 and 1.0 (3-5 hours). We then wash the cells by centrifuging at 4°C at 3000 rpm for 5 minutes and twice resuspending the cells in 25 ml ice-cold distilled water. We then centrifuge at 4°C at 3000 rpm for 5 minutes and resuspend in 1 ml ice-cold 1M sorbitol and then finally centrifuge the cells at 4°C at 5000 rpm for 5 minutes and resuspend the cells in 120 µl 1M sorbitol. To transform electrocompetent cells with plasmids or oligonucleotides, we mix 40 µl of cells with 5 µg of nucleic acid, unless otherwise stated, and incubate on ice for 5 minutes. We then transfer the mixture to a 0.2 cm electroporation cuvette and electroporate with a BIO-RAD Gene Pulser apparatus at 1.5 kV, 25 μ F, 200 Ω for one five-second pulse. We then immediately resuspend the cells in 1 ml YPD supplemented with 1M sorbitol and incubate the cultures at 30°C with shaking at 300 rpm for 6 hours. We then spread 200 µl of this culture on selective plates containing 300 µg/ml hygromycin and spread 200 µl of a 10⁵ dilution of this culture on selective plates containing 500 ng/ml aureobasidinA and/or and incubate at 30°C for 3 days to allow individual yeast colonies to grow. We then count the colonies on the plates and calculate the gene conversion efficiency by determining the number of hygromycin resistance colonies per 10⁵ aureobasidinA resistant colonies.

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Frameshift mutations are repaired in yeast cells. We test the ability of the oligonucleotides shown in Figure 8 to correct a frameshift mutation in vivo using LSY678 yeast cells

containing the plasmid pAURHYG(ins)GFP. These experiments, presented in Table 6, indicate that these oligonucleotides can support gene correction in yeast cells. These data reinforce the results described in Example 1 indicating that oligonucleotides comprising phosphorothioate linkages facilitate gene correction much more efficiently than control duplex, chimeric RNA-DNA oligonucleotides. This gene correction activity is also specific as transformation of cells with the control oligonucleotide Kan70T produced no hygromycin resistant colonies above background and thus Kan70T did not support gene correction in this system. In addition, we observe that the 74-base oligonucleotide (HygE3T/74) corrects the mutation in pAURHYG(ins)GFP approximately five-fold more efficiently than the 25-base oligonucleotide (HygE3T/25). We also perform control experiments with LSY678 yeast cells containing the plasmid pAURHYG(wt)GFP. With this strain we observed that even without added oligonucleotides, there are too many hygromycin resistant colonies to count.

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We also use additional oligonucleotides to assay the ability of individual oligonucleotides to correct multiple mutations in the pAURHYG(x)eGFP plasmid. These include, for example, one that alters two basepairs that are 3 nucleotides apart is a 74-mer with the sequence 5'-CTCGTGCTTCAGCTTCGATGTAGGAGGGCGTGGGTAACGTCCTGCGGGTAAATAGCTGCGCCGATGGTTCTAC-3'; a 74-mer that alters two basepairs that are 15 nucleotides apart with the sequence 5'-CTCGTGCTTTCAGCTTCGATGTAGGAGGGCGTGGATACGTCCTGCGGGTAAACAGCTGCGCCGATGGTTTCTAC-3'; and a 74-mer that alters two basepairs that are 27 nucleotides apart with the sequence 5'-CTCGTGCTTTCAGCTTCGATGTAGGAGGGCGTGGATACGTCCTGCGGGTAAATAGCTGCGCCGACGGTTTCTAC. The nucleotides in these oligonucleotides that direct alteration of the target sequence are underlined and in boldface. These oligonucleotides are modified in the same ways as the other oligonucleotides of the invention.

Compare the ability of single-stranded oligonucleotides to target each of the two strands of the target sequence of both pAURHYG(ins)GFP and pAURHYG(rep)GFP. These experiments, presented in Tables 7 and 8, indicate that an oligonucleotide, HygE3T/74 α , with sequence complementary to the sense strand (i.e. the strand of the target sequence that is identical to the mRNA) of the target sequence facilitates gene correction approximately ten-fold more efficiently than an oligonucleotide, HygE3T/74, with sequence complementary to the non-transcribed strand which serves as the template for the synthesis of RNA. As indicated in Table 7, this effect was observed over a range of oligonucleotide concentrations from 0-3.6 μ g, although we did observe some variability in the difference between the two oligonucleotides (indicated in Table 7 as a fold difference between HygE3T/74 α and HygE3T/74).

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Furthermore, as shown in Table 8, we observe increased efficiency of correction by HygE3T/74α relative to HygE3T/74 regardless of whether the oligonucleotides were used to correct the base substitution mutation in pAURHYG(rep)GFP or the insertion mutation in pAURHYG(ins)GFP. The data presented in Table 8 further indicate that the single-stranded oligonucleotides correct a base substitution mutation more efficiently than an insertion mutation. However, this last effect was much less pronounced and the oligonucleotides of the invention are clearly able efficiently to correct both types of mutations in yeast cells. In addition, the role of transcription is investigated using plasmids with inducible promoters such as that described in Figure 10.

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Optimization of oligonucleotide concentration. To determine the optimal concentration of oligonucleotide for the purpose of gene alteration, we test the ability of increasing concentrations of Hyg3T/74α to correct the mutation in pAURHYG(rep)GFP contained in yeast LSY678. We chose this assay system because our previous experiments indicated that it supports the highest level of correction. However, this same approach could be used to determine the optimal concentration of any given oligonucleotide. We test the ability of Hyg3T/74α to correct the mutation in pAURHYG(rep)GFP contained in yeast LSY678 over a range of oligonucleotide concentrations from 0-10.0 μg. As shown in Table 9, we observe that the correction efficiency initially increases with increasing oligonucleotide concentration, but then declines at the highest concentration tested.

Tables are attached hereto.

Table 6

Correction of an insertion mutation in pAURHYG(ins)GFP by HygGG/Rev, HygE3T/25 and HygE3T/74

Oligonucleotide Tested	Colonies on Hygromycin	Colonies on Aureobasidin (/10 ⁵)	Correction Efficiency
HygGG/Rev	3	157	0.02
HygE3T/25	64	147	0.44
HygE3T/74	280	174	1.61
Kan70T	0		

Table 7

An oligonucleotide targeting the sense strand of the target sequence corrects more efficiently.

Amount of Oligonucleotide (µg)	Colonies per hygromycin plate		
	HygE3T/74	HygE3T/74ca	
0	0	0	
0.6	24	128 (8.4x)*	
1.2	69	140 (7.5x)*	
2.4	62	167 (3.8x)*	
3.6	29	367 (15x)*	

^{*} The numbers in parentheses represent the fold increase in efficiency for targeting the non-transcribed strand as compared to the other strand of a DNA duplex that encodes a protein.

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Table 8

Correction of a base substitution mutation is more efficient than correction of a frame shift mutation.

Oligonucleotide Tested (5 µg)	Plasmid tested (contained in LSY678)	
	pAURHYG(ins)GFP	pAURHYG(rep)GFP
HygE3T/74	72	277
HygE3T/74α	1464	2248
Kan70T	0	0

Table 9

Optimization of oligonucleotide concentration in electroporated yeast cells.

Amount (µg)	Colonies on hygromycin	Colonies on aureobasidin (/10 ⁵)	Correction efficiency
0	0	67	0
1.0	5	64	0.08
2.5	47	30	1.57
5.0	199	33	6.08
7.5	383	39	9.79
10.0	191	33	5.79

Example 3 Cultured Cell Manipulation

Although disclosure in this example is directed to use of stem cells or human blood cells and microinjection, the microinjection procedures may also be used with cultured plant cells or protoplasts using any plant species, including those disclosed herein. Mononuclear cells are isolated from human umbilical cord blood of normal donors using Ficoll Hypaque (Pharmacia Biotech, Uppsala, Sweden) density centrifugation. CD34+ cells are immunomagnetically purified from mononuclear cells using either

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the progenitor or Multisort Kits (Miltenyi Biotec, Auburn, CA). Lin CD38 cells are purified from the mononuclear cells using negative selection with StemSep system according to the manufacturer's protocol (Stem Cell Technologies, Vancouver, CA). Cells used for microinjection are either freshly isolated or cryopreserved and cultured in Stem Medium (S Medium) for 2 to 5 days prior to microinjection. S Medium contains Iscoves' Modified Dulbecco's Medium without phenol red (IMDM) with 100 µg/ml glutamine/penicillin/streptomycin, 50 mg/ml bovine serum albumin, 50 µg/ml bovine pancreatic insulin, 1 mg/ml human transferrin, and IMDM; Stem Cell Technologies), 40 µg/ml low-density lipoprotein (LDL; Sigma, St. Louis, MO), 50 mM HEPEs buffer and 50 µM 2-mercaptoethanol, 20 ng/ml each of thrombopoietin, flt-3 ligand, stem cell factor and human IL-6 (Pepro Tech Inc., Rocky Hill, NJ). After microinjection, cells are detached and transferred in bulk into wells of 48 well plates for culturing.

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35 mm dishes are coated overnight at 4° C with 50 µg/ml Fibronectin (FN) fragment CH-296 (Retronectin; TaKaRa Biomedicals, Panvera, Madison, WI) in phosphate buffered saline and washed with IMDM containing glutamine/penicillin/streptomycin. 300 to 2000 cells are added to cloning rings and attached to the plates for 45 minutes at 37° C prior to microinjection. After incubation, cloning rings are removed and 2 ml of S Medium are added to each dish for microinjection. Pulled injection needles with a range of 0.22 µm to 0.3 µm outer tip diameter are used. Cells are visualized with a microscope equipped with a temperature controlled stage set at 37° C and injected using an electronically interfaced Eppendorf Micromanipulator and Transjector. Successfully injected cells are intact, alive and remain attached to the plate post injection. Molecules that are flourescently labeled allow determination of the amount of oligonucleotide delivered to the cells.

For *in vitro* erythropoiesis from Lin^CD38⁻ cells, the procedure of Malik, 1998 can be used. Cells are cultured in ME Medium for 4 days and then cultured in E Medium for 3 weeks. Erythropoiesis is evident by glycophorin A expression as well as the presence of red color representing the presence of hemoglobin in the cultured cells. The injected cells are able to retain their proliferative capacity and the ability to generate myeloid and erythoid progeny. CD34+ cells can convert a normal A (β^A) to sickle T (β^S) mutation in the β -globin gene or can be altered using any of the oligonucleotides of the invention herein for correction or alteration of a normal gene to a mutant gene. Alternatively, stem cells can be isolated from blood of humans having genetic disease mutations and the oligonucleotides of the invention can be used to correct a defect or to modify genomes within those cells.

Alternatively, non-stem cell populations of cultured cells can be manipulated using any method known to those of skill in the art including, for example, the use of polycations, cationic lipids,

liposomes, polyethylenimine (PEI), electroporation, biolistics, calcium phosphate precipitation, or any other method known in the art.

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Biolistic delivery of oligonucleotide into plant cells may be accomplished according to the following method. One milliliter of packed cell volume of plant cell suspensions are subcultured onto plates containing solid medium [with Murashige and Skoog salts from Gibco/BRL, 500 mg/liter Mes, 1 mg/liter thiamin, 100 mg/liter myo-inositol, 180 mg/liter KH2PO4, 2.21 mg/liter 2,4-dichlorophenoxyacetic acid (2,4-D), and 30 g/liter sucrose (pH 5.7) and having 8 g/liter agar-agar from Sigma added before autoclaving). By using a helium-driven particle gun such as that from BioRad and following manufacturers directions, oligonucleotides may be introduced to cells after precipitation onto 1 micrometer or comparable gold microcarriers (Bio-Rad). To precipitate onto microcarriers, 35 microliters of a particle suspension (60 mg of microcarriers per ml of 100% ethanol) is transferred to a 1.5 ml microcentrifuge tube, which is agitated on a vortex mixer. Then 40 microliter of resuspended oligonucleotide (60 ng/microliter water) is added; then 75 microliter of ice-cold 2.5 M CaCl2 is added; then 75 microliter of icecold 0.1 M spermidine is added. The tube is mixed vigorously or a vortex mixer for 10 min at room temperature. The particles are allowed to settle for 10 min and are centrifuged at 11,750 g for 30 sec. The supernatant is removed and the particles are resuspended in 50 microliter of 100% ethanol. An aliquot of 10 microliter of the resuspended particles are applied to each macro-projectile which is used to bombard each plate once at 900 psi (1 psi = 6.89 kPa) with a gap distance (distance from power source to macroprojectile) of 1 cm and a target distance (distance from microprojectile launch site to target material) of 10 cm.

An alternative method of delivery can be used as follows. Cultured cells are suspended in liquid N6 medium and then plated on a VWR Scientific glass fiber filter. About 0.4 microgram of oligonucleotide are precipitated with 15 microliter of 2.5 mM CaCl2 and 5 microliter of 0.1 M spermidine onto 25 microgram of 1.0 micrometer gold particles. Microprojectile bombardment is performed by using a Bio-Rad PDS-1000 He particle delivery system or comparable machine following manufacturers instructions. Alterations in oligonucleotide concentrations can be employed to determine the optimum concentration of oligonucleotide according to the procedures described herein for any particular oligonucleotide of the invention.

Alternatively, the oligonucleotide of the invention may be delivered to a plant cell by electroporation of a protoplast derived from a plant part. The protoplasts may be formed by enzymatic treatment of a plant part, particularly a leaf, according to techniques such as those in Gallois et al., Methods in Molecular Biology 55: 89-107 by Humana Press. Such conditions for electroporation use

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about 3×10^5 protoplasts in a total volume of about 0.3 ml with a concentration of oligonucleotide of between 0.6 to 4 microgram per ml.

EXAMPLE 4

Plant Cells

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event in plants and animal cells. Although little information is available on plant mutations amongst natural cultivars, the oligonucleotides of the invention can be used to produce "knock out" mutations by modification of specific amino acid codons to produce stop codons (e.g., a CAA codon specifying Gln can be modified at a specific site to TAA; a AAG codon specifying Lys can be modified to UAG at a specific site; and a CGA codon for Arg can be modified to a UGA codon at a specific site). Such base pair changes will terminate the reading frame and produce a defective truncated protein, shortened at the site of the stop codon

. Alternatively, frameshift additions or deletions can be directed into the genome at a specific sequence to interrupt the reading frame and produce a garbled downstream protein. Such stop or frameshift mutations can be introduced to determine the effect of knocking out the protein in either plant or animal cells.

For introduction of a T-DNA, including the T-DNA in the plasmid of Figure 11, into a plant cell, *Agrobacterium tumefaciens* is used. These techniques are routine standard techniques known in the art. For example, one method follows. We transform *A. tumefaciens* is transformed by electroporation (using a BioRad Gene Pulser^{\mathbb{I}}). Competent *A. tumefaciens* is prepared using a method similar to that of preparing competent *E. coli* by suspending a freshly grown culture three times in ice-cold water and a final resuspension in 10% glycerol. Electroporation conditions are a 0.2 cm gap cuvette at a setting of 25 μ F, 200 Ω and 2.5 kV.

A tumefaciens containing a plasmid with a T-DNA is then used to introduce the T-DNA into a plant cell using routine standard techniques known in the art. For example, we transform Arabidopsis by vacuum infiltration or by dipping flowers in an Agrobacterium solution containing a surfactant, e.g. L-77. Seeds are then collected, grown and screened for presence of the T-DNA. Alternatively, Agrobacterium can be used to transform callus tissue and the callus tissue can then be used to regenerate transformed plants.

All publications and patent applications cited in this specification are herein incorporated by reference as if each individual publication or patent application were specifically and individually indicated to be incorporated by reference. Although the foregoing invention has been described in some

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detail by way of illustration and example for purposes of clarity of understanding, it will be readily apparent to those of ordinary skill in the art in light of the teachings of this invention that certain changes and modifications may be made thereto without departing from the spirit or scope of the appended claims.

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Notes on the tables presented below:

Each of the following tables presents, for the specified gene, a plurality of mutations that are known to confer a relevant phenotype and, for each mutation, the oligonucleotides that can be used to correct the respective mutation site-specifically in the genome according to the present invention.

The left-most column identifies each alteration or mutation and the phenotype that the alteration/mutation confers.

For most entries, the mutation/alteration is identified at both the nucleic acid and protein level. At the amino acid level, mutations are presented according to the following standard nomenclature. The centered number identifies the position of the mutated codon in the protein sequence; to the left of the number is the wild type residue and to the right of the number is the mutant codon. Terminator codons are shown as "TERM". At the nucleic acid level, the entire triplet of the wild type and mutated codons is shown.

The middle column presents, for each mutation, four oligonucleotides capable of repairing the mutation site-specifically in the genome or in cloned DNA including DNA in artificial chromosomes, episomes, plasmids, or other types of vectors. The oligonucleotides of the invention, however, may include any of the oligonucleotides sharing portions of the sequence of the 121 base sequence. Thus, oligonucleotides of the invention for each of the depicted targets may be 18, 19, 20 up to about 121 nucleotides in length. Sequence may be added non-symmetrically.

All oligonucleotides are presented, per convention, in the 5' to 3' orientation. The nucleotide that effects the change in the genome is underlined and presented in bold.

The first of the four oligonucleotides for each mutation is a 121 nt oligonucleotide centered about the repair/altering nucleotide. The second oligonucleotide, its reverse complement, targets the opposite strand of the DNA duplex for repair/alteration. The third oligonucleotide is the minimal 17 nt domain of the first oligonucleotide, also centered about the repair/alteration nucleotide. The fourth oligonucleotide is the reverse complement of the third, and thus represents the minimal 17 nt domain of the second.

The third column of each table presents the SEQ ID NO: of the respective repair oligonucleotide.

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Example 5

Engineering herbicide resistant plants

Chemical weed control is an important tool of modern agriculture and many herbicides have been developed for this purpose. Their use has resulted in substantial increases in the yields of many crops, including, for example, maize, soybeans, and cotton. Thus while the use of fertilizers and new high-yielding crop varieties have contributed greatly to the "green revolution," chemical weed control has also been at the forefront of technological achievement.

Herbicides having broad-spectrum activity are particularly useful because they obviate the need for multiple herbicides targeting different classes of weeds. The problem with such herbicides is that they typically also affect crops which are exposed to the herbicide. One way to overcome this is to generate plants which are resistant to one or more broad-spectrum herbicides. Such herbicide-tolerant plants may reduce the need for tillage to control weeds, thereby effectively reducing soil erosion and can reduce the quantity and number of different herbicides applied in the field.

Common herbicides used, for example, include those that inhibit the enzyme 5-enolpyruvyl-3-phosphoshikimic acid synthase (EPSPS), for example N-phosphonomethyl-glycine (e.g. glyphosate), those that inhibit acetolactate synthase (ALS) activity, for example the sulfonylureas and related herbicides, and those that inhibit dihydropteroate synthase, for example methyl[(4-aminophenyl)sulfonyl]carbamate (e.g. Asulam). Herbicide-tolerant plants can be produced by several methods, including, for example, introducing into the genome of the plant the ability to degrade the herbicide, the capacity to produce a higher level of the targeted enzyme, and/or expressing an herbicide-tolerant allele of the enzyme.

The attached tables disclose exemplary oligonucleotides base sequences which can be used to generate site-specific mutations in plant genes that confer herbicide resistance.

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Table 10
Genome-Altering Oligos Conferring Glyphosate Resistance

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Glyphosate Resistance EPSPS <i>Arabidopsis thaliana</i>	AAGCGTCGGAGATTGTACTTCAACCCATTAGAGAAATCTCCGGTC TTATTAAGCTTCCTG <u>C</u> CTCCAAGTCTCTATCAAATCGGATCCTGC TTCTCGCTGCTCTGTCTGAGGTATATATCAC	4341
Gly97Ala GGC-GCC	GTGATATACCTCAGACAGAGCAGCGAGAAGCAGGATCCGATT TGATAGAGACTTGGAGGCAGGAAGCTTAATAAGACCGGAGATTT CTCTAATGGGTTGAAGTACAATCTCCGACGCTT	4342
	GCTTCCTG <u>C</u> CTCCAAGT	4343
	ACTTGGAG <u>G</u> CAGGAAGC	4344
Glyphosate Resistance EPSPS Brassica napus	AAGCTTCAGAGATTGTGCTTCAACCAATCAGAGAAATCTCGGGTC TCATTAAGCTACCCGCATCCAAATCTCTCCCAATCGGATCCTCC TTCTTGCCGCTCTATCTGAGGTACATATACT	4345
Gly93Ala GGA-GCA	AGTATATGTACCTCAGATAGAGCGGCAAGAAGGAGGATCCGATT GGAGAGAGATTTGGATGCGGGTAGCTTAATGAGACCCGAGATTT CTCTGATTGGTTGAAGCACAATCTCTGAAGCTT	4346
	GCTACCCG <u>C</u> ATCCAAAT	4347
	ATTTGGAT <u>G</u> CGGGTAGC	4348
Glyphosate Resistance EPSPS 1 Nicotiana tabacum	AGCCCAACGAGATTGTGCTGCAACCCATCAAAGATATATCAGGC ACTGTTAAATTGCCTGCTTCTAAATCCCTTTCCAATCGTATTCTCC TTCTTGCTGCCCTTTCTAAGGGAAGGACTGT	4349
Gly95Ala GGT-GCT	ACAGTCCTTCCCTTAGAAAGGGCAGCAAGAAGGAGAATACGATT GGAAAGGGATTTAGAAGCAGGCAATTTAACAGTGCCTGATATATC TTTGATGGGTTGCAGCACAATCTCGTTGGGCT	4350
	ATTGCCTG <u>C</u> TTCTAAAT	4351
	ATTTAGAA <u>G</u> CAGGCAAT	4352
Glyphosate Resistance EPSPS 2 Nicotiana tabacum	ATTGTTTCCTTGGTACGAAATGTCCTCCTGTTCGAATTGTCAGCA AGGGAGGCCTTCCCGCAGGGAAGGTAAAGCTCTCTGGATCAATT AGCAGCCAGTACTTGACTGCTCTGCT	4353
Gly62Ala GGA-GCA	GCCATAAGCAGAGCAGTCAAGTACTGGCTGCTAATTGATCCAGA GAGCTTTACCTTCCCTGCGGGAAGGCCTCCCTTGCTGACAATTC GAACAGGAGGACATTTCGTACCAAGGAAACAAT	4354
	CCTTCCCG <u>C</u> AGGGAAGG	4355
	CCTTCCCT <u>G</u> CGGGAAGG	4356
Glyphosate Resistance EPSPS Zea mays	ATTGTTTCCTTGGCACTGACTGCCCACCTGTTCGTGTCAATGGAA TCGGAGGGCTACCTGCTGGCAAGGTCAAGCTGTCTGGCTCCATC AGCAGTCAGTACTTGAGTGCCTTGCTGATGGC	4357
Gly168Ala GGT-GCT	GCCATCAGCAAGGCACTCAAGTACTGACTGCTGATGGAGCCAGA CAGCTTGACCTTGCCAGCAGGTAGCCCTCCGATTCCATTGACAC GAACAGGTGGGCAGTCAGTGCCAAGGAAACAAT	4358

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Aiteration	GCTACCTGCTGGCAAGG	4359
	CCTTGCCAGCAGGTAGC	4360
Glyphosate Resistance EPSPS	ACTGTTTCCTTGGCACTGAATGCCCACCTGTTCGTGTCAAGGGA ATTGGAGGACTTCCTGCTGGCAAGGTTAAGCTCTCTGGTTCCAT CAGCAGTCAGTACTTGAGTGCCTTGCTGATGGC	4361
Oryza sativa Gly115Ala GGT-GCT	GCCATCAGTACTTGAGTGCCTTCGTCTTGATGGAACCAGA GCCATCAGCAAGGCACTCAAGTACTGACTGCTGATGGAACCAGA GAGCTTAACCTTGCCAGCAGGAAGTCCTCCAATTCCCTTGACAC GAACAGGTGGGCATTCAGTGCCAAGGAAACAGT	4362
	ACTTCCTGCTGGCAAGG	4363
•	CCTTGCCAGCAGGAAGT	4364
Glyphosate Resistance EPSPS	AGCCTTCTGAGATAGTGTTGCAACCCATTAAAGAGATTTCAGGCA CTGTTAAATTGCCTGCCTCTAAATCATTATCTAATAGAATTCTCCT TCTTGCTGCCTTATCTGAAGGAACAACTGT	4365
<i>Petunia x hybrida</i> Gly93Ala GGC-GCC	ACAGTTGTTCCTTCAGATAAGGCAGCAAGAAGGAGAATTCTATTA GATAATGATTTAGAGGCAGCAATTTAACAGTGCCTGAAATCTCT TTAATGGGTTGCAACACTATCTCAGAAGGCT	4366
	ATTGCCTG <u>C</u> CTCTAAAT	4367
	ATTTAGAGGCAGGCAAT	4368
Glyphosate Resistance EPSPS	TOTAL CONTRACTOR AND A TATAL CTCCTA	4369
Lycopersicon esculentum Gly97Ala	ACAGTCCTTCCGAGAAAGGGCAGCAAGAAGGAGAATACGATT GGAAAGGGATTTCGAAGCGGGTAATTTAACAGTACCAGATATATC TTTGATGGTNCTAGCACAATCTCATGGGGTT	437
GGT-GCT	ATTACCCG <u>C</u> TTCGAAAT	437
	ATTTCGAA <u>G</u> CGGGTAAT	437
Glyphosate Resistance	E ATTGTTTCCTTGGCACTGACTGCCCACCTGTTCGKATCAACGGCA TTGGAGGGCTACCTGCTGGCAAGGTTAAGCTGTCCATC AGCAGCCAATACTTGAGTTCCTTGCTGATGGC	437
Lolium rigidum Gly107Ala GGT-GCT	GCCATCAGCAAGGAACTCAAGTATTGGCTGCTGATGGAACCAGA CAGCTTAACCTTGCCAGCAGGTAGCCCTCCAATGCCGTTGATCG AACAGGTGGGCAGTCAGTGCCAAGGAAACAAT	437
	GCTACCTGCTGGCAAGG	437
	CCTTGCCAGCAGGTAGC	43

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Table 11

<u>Genome-Altering Oligos Conferring Imidazolinone and Sulfonylurea Herbicide Resistance</u>

5	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Sulfonylurea Resistance ALS	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCTCTTGTAGCA ATCACAGGACAAGTC <u>T</u> CTCGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4377
10	Arabidopsis thaliana Pro197Ser CCT-TCT	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACGAGAGGACTTGTCCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4378
		GACAAGTC <u>T</u> CTCGTCGT	4379
		ACGACGAG <u>A</u> GACTTGTC	4380
	Sulfonylurea Resistance ALS	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCTCTTGTAGCA ATCACAGGACAAGTCCAGCGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4381
15	Arabidopsis thaliana Pro197Gln CCT-CAG	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACG <u>CT</u> GGACTTGTCCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4382
		ACAAGTCC <u>AG</u> CGTCGTC	4383
		TACGACG <u>CT</u> GGACTTGT	4384
20	Sulfonylurea Resistance ALS	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCTCTTGTAGCA ATCACAGGACAAGTCC <u>AA</u> CGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4385
	Arabidopsis thaliana Pro197Gln CCT-CAA	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACG <u>TT</u> GGACTTGTCCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4386
		ACAAGTCC <u>AA</u> CGTCGTA	4387
`		TACGACG <u>TT</u> GGACTTGT	4388
25	Imidazolinone Resistance ALS	GACCTTACCTGTTGGATGTGATTTGTCCGCACCAAGAACATGTGT TGCCGATGATCCCGA <u>AC</u> GGTGGCACTTTCAACGATGTCATAACGG AAGGAGATGGCCGGATTAAATACTGAGAGAT	4389
	Arabidopsis thaliana Ser653Asn AGT-AAC	ATCTCTCAGTATTTAATCCGGCCATCTCCTTCCGTTATGACATCGT TGAAAGTGCCACC <u>GT</u> TCGGGATCATCGGCAACACATGTTCTTGGT GCGGACAAATCACATCCAACAGGTAAGGTC	4390
		GATCCCGA <u>AC</u> GGTGGCA	4391
		TGCCACC <u>GT</u> TCGGGATC	4392

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Imidazolinone Resistance	GACCTTACCTGTTGGATGTGATTTGTCCGCACCAAGAACATGTGT TGCCGATGATCCCGAATGGTGGCACTTTCAACGATGTCATAACGG AAGGAGATGGCCGGATTAAATACTGAGAGAT	4393
ALS Arabidopsis thaliana Ser653Asn	ATCTCTCAGTATTTAATCCGGCCATCTCCTTCCGTTATGACATCGT TGAAAGTGCCACCATTCGGGATCATCGGCAACACATGTTCTTGGT GCGGACAAATCACATCCAACAGGTAAGGTC	4394
AGT-AAT	GATCCCGA <u>AT</u> GGTGGCA	4395
-	TGCCACC <u>AT</u> TCGGGATC	4396
Sulfonylurea Resistance	TCCGCGCTCGCCGACGCGCTGCTCGACTCCGATGGTCG CCATCACGGGCCAGGTCTCCCGCCGCATGATCGGCACCGC CTTCCAGGAGACGCCCATAGTCGAGGTCACCCGCT	4397
ALS Oryza sativa Pro171Ser	AGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGTG CCGATCATGCGGCGGGAGACCTGGCCCGTGATGGCGACCATCG GGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGA	4398 ,
CCC-TCC	GCCAGGTCTCCCGCCGC	4399
	GCGGCGG <u>A</u> GACCTGGC	4400
Sulfonylurea Resistance	CCGCGCTCGCCGACGCGCTGCTCGACTCCGATGGTCGC CATCACGGGCCAGGTCCAACGCCGCATGATCGGCACCGACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4401
ALS Oryza sativa Pro171Gln	GAGCGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGTTGGACCTGGCCCGTGATGGCGACCATCG	4402
CCC-CAA	GGACGGAGTCGAGCGCGCGCGAGCGCGG CCAGGTCCAACGCCGCA	4403
	TGCGGCG <u>TT</u> GGACCTGG	4404
Sulfonylurea Resistance	CCGCGCTCGCCGACGCGCTGCTCGACTCCGATGGTCGC CATCACGGGCCAGGTCCAGCGCCCATGATCGGCACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4405
ALS Oryza sativa Pro171Gln	GAGCGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGCTGGACCTGGCCCGTGATGGCGACCATC GGGACGGAGTCGAGCAGCGCGCGAGCGCGG	4406
CCC-CAG	CCAGGTCCAGCGCGCA	440
	TGCGGCG <u>CT</u> GGACCTGG	4408
Imidazolinone Resistance	GGCCATACTTGTTGGATATCATCGTCCCGCACCAGGAGCATGTGC TGCCTATGATCCCAAATGGGGGCGCATTCAAGGACATGATCCTGG ATGGTGATGGCAGGACTGTGTATTAATCTAT	440
ALS Oryza sativa Ile627Asn	ATAGATTAATACACAGTCCTGCCATCACCATCCAGGATCATGTCCT TGAATGCGCCCCCATTTGGGATCATAGGCAGCACATGCTCCTGGT	441
ATT-AAT	GCGGGACGATGATATCCAACAAGTATGGCC GATCCCAAATGGGGGCG	441

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQIC NO:
	CGCCCCATTTGGGATC	4412
Sulfonylurea Resistance ALS	TCCGCGCTCGCCGACGCGCTGCTCGATTCCGTCCCCATGGTCGC CATCACGGGACAGGTGTCGCGACGCATGATTGGCACCGACGCCT TCCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4413
Zea mays Pro165Ser CCG-TCG	AGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCGGT GCCAATCATGCGTCGCGACACCTGTCCCGTGATGGCGACCATGG GGACGGAATCGAGCAGCGCGTCGGCGAGCGCGGA	4414
	GACAGGTG <u>T</u> CGCGACGC	4415
•	GCGTCGCG <u>A</u> CACCTGTC	4416
Sulfonylurea Resistance ALS	CCGCGCTCGCCGACGCGCTGCTCGATTCCGTCCCCATGGTCGCC ATCACGGGACAGGTGCAGCGACGCATGATTGGCACCGACGCCTT CCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4417
Zea mays Pro165Gln CCG-CAG	GAGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCGG TGCCAATCATGCGTCGCTGCACCTGTCCCGTGATGGCGACCATG GGGACGGAATCGAGCAGCGCGTCGGCGAGCGCGG	4418
	ACAGGTGC <u>A</u> GCGACGCA	4419
	TGCGTCGC <u>T</u> GCACCTGT	4420
Imidazolinone Resistance ALS	GGCCGTACCTCTTGGATATAATCGTCCCACACCAGGAGCATGTGT TGCCTATGATCCCTAATGGTGGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTACTGATCTAA	4421
Zea mays Ser621Asn AGT-AAT	TTAGATCAGTACACAGTCCTGCCATCACCATCCAGGATCATATCCT TGAAAGCCCCACCATTAGGGATCATAGGCAACACATGCTCCTGGT GTGGGACGATTATATCCAAGAGGTACGGCC	4422
	GATCCCTA <u>AT</u> GGTGGGG	4423
	CCCCACC <u>AT</u> TAGGGATC	4424
Imidazolinone Resistance ALS	GGCCGTACCTCTTGGATATAATCGTCCCACACCAGGAGCATGTGT TGCCTATGATCCCTAACGGTGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTACTGATCTAA	4425
Zea mays Ser621Asn AGT-AAC	TTAGATCAGTACACAGTCCTGCCATCACCATCCAGGATCATATCCT TGAAAGCCCCACCGTTAGGGATCATAGGCAACACATGCTCCTGGT GTGGGACGATTATATCCAAGAGGTACGGCC	4426
	GATCCCTA <u>AC</u> GGTGGGG	4427
	CCCCACC <u>GT</u> TAGGGATC	4428
Sulfonylurea Resistance ALS	TCCGCGCTCGCCGACGCCCTCCTCGACTCCATCCCCATGGTGGC CATCACGGGGCAGGTCTCGCGCCGCATGATCGGCACGGACGCC TTCCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4429
Lolium multiflorum Pro167Ser CCG-TCG	AGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCCGT GCCGATCATGCGGCGCGAGACCTGCCCCGTGATGGCCACCATG GGGATGGAGTCGAGGAGGGCGTCGCCGAGCGCGGA	4430

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GGCAGGTC <u>T</u> CGCGCCGC	4431
	GCGGCGCGAGACCTGCC	4432
Sulfonylurea Resistance	CCGCGCTCGCCGACGCCCTCCTCGACTCCATCCCCATGGTGGCC ATCACGGGGCAGGTCCAGCGCCGCATGATCGGCACGGACGCCT TCCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4433
ALS L <i>olium multiflorum</i> Pro167GIn	GAGCGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCCG TGCCGATCATGCGGCGCTGGACCTGCCCCGTGATGGCCACCATG	4434
CCG-CAG	GGGATGGAGTCGAGGGGGGGGGGGGGGGGGGGGGGGGGG	4435
	TGCGGCGC <u>T</u> GGACCTGC	4436
Imidazolinone Resistance	CTGGGCCATACTTGTTGGATATCATCGTCCCTCACCAGGAGCATG TGCTGCCTATGATCCCTAACGGTGGTGCTTTCAAGGACATTATCA	4437
ALS Lolium multiflorum Ser623Asn	TGGAAGGTGATGGCAGGATTTCGTATTAAAC GTTTAATACGAAATCCTGCCATCACCTTCCATGATAATGTCCTTGA AAGCACCACCGTTAGGGATCATAGGCAGCACATGCTCCTGGTGA GGGACGATGATATCCAACAAGTATGGCCCAG	4438
AGC-AAC	GGGACGATGATATCCAACAAGTATCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCACAAGTATCCCCCCACAAGTATCCCCCCACAAGTATCCCCCCAACAAGTATCCCCCAACAAGTATCCCCCAACAAGTATCCCCCAACAAGTATCCCCCCAACAAGTATCCCCCCAACAAGTATCCCCCCAACAAGTATCCCCCCAACAAGTATCCCCCAACAAAGTATCCCCCAACAAGTATCCCCCAACAAGTATCCCCAACAAGTATCCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCCAACAAGTATCCCAACAAGTATCCCAACAAGTATCCCAACAAGTATCCCAACAAGTATCCCAACAAAGTATCCCAACAAAGTATCCCAACAAAGTATCCCAACAAAGTATCCCAACAAAGTATCCCAACAAAGTATCCCAACAAAAGTATCCCAACAAAAAAAA	4439
	CACCACCGTTAGGGATC	4440
Sulfonylurea Resistance	TCCGCGCTCGCCGACGCTCTCCTCGACTCCATCCCCATGGTCGC CATCACGGGCCAGGTCTCACGCCGCATGATCGGCACGCGCT	4441
ALS Hordeum vulgare Pro68Ser	TCCAGGAGACGCCCATAGTGGAGGTCACGCGCT AGCGCGTGACCTCCACTATGGGCGTCTCCTGGAACGCGTCCGTG CCGATCATGCGGCGTGAGACCCCGAGCGCGCGACCATGG	4442
CCA-TCA	GGATGGAGTCGAGGAGGGGGGGAGCGCGGAGCGCGGGAGCGCGGGAGCGCGGCG	444
	GCGGCGTG <u>A</u> GACCTGGC	444
Sulfonylurea Resistance	CCGCGCTCGCCGACGCTCTCCTCGACTCCCATCCCCATGGTCGCC ATCACGGCCCAGGTCCAACGCCGCATGATCGGCACGGACGCGTT CCAGGAGACGCCCATAGTGGAGGTCACGCGCTC	444
ALS Hordeum vulgare Pro68Gin	GAGCGCGTGACCTCCACTATGGGCGTCTCCTGGAACGCGTCCGT GCCGATCATGCGGCGTTGGACCTGGCCCGTGATGGCGACCATGG GCATGGAGTCGAGGAGAGCGTCGGCGAGCGCGG	444
CCA-CAA	CCAGGTCCAACGCCGCAACCCCCC	444
	TGCGGCGT <u>T</u> GGACCTGG	444

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ NO:
lmidazolinone Resistance ALS	CCCAGGGCCGTACCTGCTGGATATCATTGTCCCGCATCAGGAGC ACGTGCTGCCTATGATCCCAAACGGTGGTGCTTTCAAGGACATGA TCATGGAGGGTGATGGCAGGACCTCGTACTGA	444!
<i>Hordeum vulgare</i> Ser524Asn AGC-AAC	TCAGTACGAGGTCCTGCCATCACCCTCCATGATCATGTCCTTGAA AGCACCACCGTTTGGGATCATAGGCAGCACGTGCTCCTGATGCG GGACAATGATATCCAGCAGGTACGGCCCTGGG	445
	GATCCCAA <u>A</u> CGGTGGTG	445
	CACCACCG <u>T</u> TTGGGATC	445
Sulfonylurea Resistance ALS	AGTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCG ATCACTGGTCAAGTCTCTCGTCGGATGATCGGTACCGATGCTTTC CAGGAAACTCCAATTGTTGAGGTAACAAGGT	445
Gossypium hirsutum Pro186Ser CCT-TCT	ACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTAC CGATCATCCGACGAGAGAGCGGACCACGAGAGAGGG ATACTATCGAGCATTGCATCAGCGAGACCACT	445
	GTCAAGTC <u>T</u> CTCGTCGG	445
_	CCGACGAG <u>A</u> GACTTGAC	445
Sulfonylurea Resistance ALS	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCC <u>AA</u> CGTCGGATGATCGGTACCGATGCTTTCC AGGAAACTCCAATTGTTGAGGTAACAAGGTC	445
Gossypium hirsutum Pro186Gln CCT-CAA	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACG <u>TT</u> GGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	445
	TCAAGTCC <u>AA</u> CGTCGGA	445
	TCCGACG <u>TT</u> GGACTTGA	446
Sulfonylurea Resistance ALS	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCAGCGTCGGATGATCGGTACCGATGCTTTCC AGGAAACTCCAATTGTTGAGGTAACAAGGTC	446
Gossypium hirsutum Pro186Gln CCT-CAG	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACG <u>CT</u> GGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	446
	TCAAGTCC <u>AG</u> CGTCGGA	446
	TCCGACG <u>CT</u> GGACTTGA	446
Imidazolinone Resistance ALS	GACCTTACTTGTTGGATGTGATTGTCCCACATCAAGAACATGTCCT GCCTATGATCCCCAATGGGGGGGCGCTTTCAAAGATGTGATCACAGA GGGTGATGGAAGAACACAATATTGACCTCA	446
Gossypium hirsutum Ser642Asn AGT-AAT	TGAGGTCAATATTGTGTTCTTCCATCACCCTCTGTGATCACATCTT TGAAAGCGCCTCCATTGGGGATCATAGGCAGGACATGTTCTTGAT GTGGGACAATCACATCCAACAAGTAAGGTC	446
	GATCCCCA <u>A</u> TGGAGGCG	446

	Phenotype: Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	Alteration	CGCCTCCA <u>T</u> TGGGGATC	4468
	Sulfonylurea Resistance	TCTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCC ATTACTGGGCAAGTTTCCCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACTCGAT	4469
	ALS Amaranthus retroflexus	ATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTACC AATCATACGCCGGGAAACTTGCCCAGTAATGGCGACAAGAGGGA	4470
	Pro192Ser CCC-TCC	CTGAGTCAAGAAGTGCATCAGCAAGACCAGA GGCAAGTT <u>T</u> CCCGGCGT	4471
		ACGCCGGG <u>A</u> AACTTGCC	4472
	Sulfonylurea Resistance	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAACGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACTCGATC	4473
	ALS Amaranthus retroflexus	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGTTGAACTTGCCCAGTAATGGCGACAAGAGGGA CTGAGTCAAGAAGTGCATCAGCAAGACCAG	4474
	Pro192Gln CCC-CAA	GCAAGTTC <u>AA</u> CGGCGTA	4475
		TACGCCG <u>TT</u> GAACTTGC	4476
	Sulfonylurea Resistance	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAGCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACTCGATC	4477
	ALS Amaranthus retroflexus	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGCTGAACTTGCCCAGTAATGGCGACAAGAGGG ACTGAGTCAAGAAGTGCATCAGCAAGACCAG	4478
	Pro192Gln CCC-CAG	GCAAGTTC <u>AG</u> CGGCGTA	447
		TACGCCG <u>CT</u> GAACTTGC	448
	Imidazolinone Resistance	GACCGTATCTGCTGGATGTAATCGTACCACATCAGGAGCATGTGC TGCCTATGATCCCTAACGGTGCCGCCTTCAAGGACACCATAACAG AGGGTGATGGAAGAAGGGCTTATTAGTTGGT	<u>' </u>
5	ALS Amaranthus retroflexus	ACCAACTAATAAGCCCTTCTTCCATCACCCTCTGTTATGGTGTCCT TGAAGGCGGCACCGTTAGGGATCATAGGCAGCACATGCTCCTGA TGTGGTACGATTACATCCAGCAGATACGGTC	448
	Ser652Asn AGC-AAC	GATCCCTAACGGTGCCG	448
		CGGCACCGTTAGGGATC	448

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Sulfonylurea Resistance ALS 1	AGCGGCCTCGCTGACGCGCTACTGGATAGCGTCCCCATTGTTGC TATAACAGGTCAAGTGTCACGTAGGATGATAGGTACTGATGCTTTT CAGGAAACTCCTATTGTTGAGGTAACTAGAT	4485
5	Nicotiana tabacum Pro194Ser CCA-TCA	ATCTAGTTACCTCAACAATAGGAGTTTCCTGAAAAGCATCAGTACC TATCATCCTACGTGACACTTGACCTGTTATAGCAACAATGGGGAC GCTATCCAGTAGCGCGTCAGCGAGGCCGCT	4486
		GTCAAGTG <u>T</u> CACGTAGG	4487
	`	CCTACGTG <u>A</u> CACTTGAC	4488
	Sulfonylurea Resistance ALS 1	GCGGCCTCGCTGACGCGCTACTGGATAGCGTCCCCATTGTTGCT ATAACAGGTCAAGTGCAACGTAGGATGATAGGTACTGATGCTTTT CAGGAAACTCCTATTGTTGAGGTAACTAGATC	4489
10	Nicotiana tabacum Pro194Gln CCA-CAA	GATCTAGTTACCTCAACAATAGGAGTTTCCTGAAAAGCATCAGTAC CTATCATCCTACGT <u>T</u> GCACTTGACCTGTTATAGCAACAATGGGGA CGCTATCCAGTAGCGCGTCAGCGAGGCCGC	4490
		TCAAGTGC <u>A</u> ACGTAGGA	4491
		TCCTACGT <u>T</u> GCACTTGA	4492
15	Imidazolinone Resistance ALS 1	GGCCATACTTGTTGGATGTGATTGTACCTCATCAGGAACATGTTTT ACCTATGATTCCCAATGGCGGAGCTTTCAAAGATGTGATCACAGA GGGTGACGGGAGAAGTTCCTATTGAGTTTG	4493
	Nicotiana tabacum Ser650Asn AGT-AAT	CAAACTCAATAGGAACTTCTCCCGTCACCCTCTGTGATCACATCTT TGAAAGCTCCGCCATTGGGAATCATAGGTAAAACATGTTCCTGAT GAGGTACAATCACATCCAACAAGTATGGCC	4494
•		GATTCCCA <u>A</u> TGGCGGAG	4495
		CTCCGCCATTGGGAATC	4496
20	Sulfonylurea Resistance ALS 2	AGTGGCCTCGCGGACGCCCTACTGGATAGCGTCCCCATTGTTGC TATAACCGGTCAAGTGTCACGTAGGATGATCGGTACTGATGCTTTT CAGGAAACTCCGATTGTTGAGGTAACTAGAT	4497
	Nicotiana tabacum Pro191Ser CCA-TCA	ATCTAGTTACCTCAACAATCGGAGTTTCCTGAAAAGCATCAGTACC GATCATCCTACGTGACACTTGACCGGTTATAGCAACAATGGGGAC GCTATCCAGTAGGGCGTCCGCGAGGCCACT	4498
		GTCAAGTG <u>T</u> CACGTAGG	4499
		CCTACGTG <u>A</u> CACTTGAC	4500
25	Sulfonylurea Resistance ALS 2	GTGGCCTCGCGGACGCCCTACTGGATAGCGTCCCCATTGTTGCT ATAACCGGTCAAGTGCAACGTAGGATGATCGGTACTGATGCTTTT CAGGAAACTCCGATTGTTGAGGTAACTAGATC	4501
30	Nicotiana tabacum Pro191Gln CCA-CAA	GATCTAGTTACCTCAACAATCGGAGTTTCCTGAAAAGCATCAGTAC CGATCATCCTACGTTGCACTTGACCGGTTATAGCAACAATGGGGA CGCTATCCAGTAGGGCGTCCGCGAGGCCAC	4502
	20	TCAAGTGC <u>A</u> ACGTAGGA	4503

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Aileration	TCCTACGT <u>T</u> GCACTTGA	4504
Imidazolinone Resistance	GGCCATACTTGTTGGATGTGATTGTACCTCATCAGGAACATGTTCT ACCTATGATTCCCAATGGCGGGGCTTTCAAAGATGTGATCACAGA GGGTGACGGGAGAAGTTCCTATTGACTTTG	4505
ALS 2 Nicotiana tabacum Ser647Asn	CAAAGTCAATAGGAACTTCTCCCGTCACCCTCTGTGATCACATCTT TGAAAGCCCCGCCATTGGGAATCATAGGTAGAACATGTTCCTGAT GAGGTACAATCACATCCAACAAGTATGGCC	4506
AGT-AAT	GATTCCCAATGCGGGG	4507
	CCCCGCCATTGGGAATC	4508
Sulfonylurea Resistance	AGTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTA TTACTGGTCAAGTTTCCAGGAGAATGATTGGAACAGATGCGTTTCA	4509
ALS Xanthium spp. Pro175Ser	AACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTCC AATCATTCTCCTGGAAACTTGACCAGTAATAGCAACCATTGGAACA CTGTCTAATAAAGCATCAGCAAGACCACT	4510
CCC-TCC	GTCAAGTT <u>T</u> CCAGGAGA	4511
	TCTCCTGG <u>A</u> AACTTGAC	4512
Sulfonylurea Resistance	GTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTAT TACTGGTCAAGTTCAAAGGAGAGAATGATTGGAACAGATGCGTTTCA AGAAACCCCTATTGTTGAGGTAACACGTTC	4513
ALS Xanthium spp. Pro175Gln	GAACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTC CAATCATTCTCCT <u>TT</u> GAACTTGACCAGTAATAGCAACCATTGGAAC ACTGTCTAATAAAGCATCAGCAAGACCAC	4514
CCC-CAA	TCAAGTTC <u>AA</u> AGGAGAA	451
	TTCTCCT <u>TT</u> GAACTTGA	451
Sulfonylurea Resistance	GTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTAT TACTGGTCAAGTTCAGAGGAGAATGATTGGAACAGATGCGTTTCA AGAAACCCCTATTGTTGAGGTAACACGTTC	
ALS Xanthium spp. Pro175Gln	GAACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTC CAATCATTCTCCTCTGAACTTGACCAGTAATAGCAACCATTGGAAC ACTGTCTAATAAAGCATCAGCAAGACCAC	451
CCC-CAG	TCAAGTTCAGCAGCACAGCAGCAGCAGCAGCAGGAGAGAGA	451
	TTCTCCT <u>CT</u> GAACTTGA	452
Imidazolinone Resistance	GGGCCTTACTTGTTGGATGTGATCGTGCCCCATCAAGAACATGTG TTGCCCATGATCCCGAATGGTGGAGGTTTCATGGATGTGATCACC GAAGGCGACGCAGAATGAAATATTGAGCTT	
ALS Xanthium spp. Ala631Asn	AAGCTCAATATTTCATTCTGCCGTCGCCTTCGGTGATCACATCCAT GAAACCTCCACCATTCGGGATCATGGGCAACACATGTTCTTGATG	452
GCT-AAT	GGGCACGATCACATCCAACAAGTAAGGCCC	

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	TGATCCCG <u>AA</u> TGGTGGA	4523
	TCCACCA <u>TT</u> CGGGATCA	4524
Sulfonylurea Resistance ALS	TCCGGGTTTGCTGATGCTTTGCTCGATTCCGTTCCACTGGTGGCG ATCACGGGGCAGGTGTCGCGGCGAATGATTGGGACGGATGCTTT TCAGGAGACTCCTATTGTTGAGGTAACACGGT	4525
Bassia scoparia Pro189Ser CCG-TCG	ACCGTGTTACCTCAACAATAGGAGTCTCCTGAAAAGCATCCGTCC CAATCATTCGCCGCGACACCTGCCCCGTGATCGCCACCAGTGGA ACGGAATCGAGCAAAGCATCAGCAAACCCGGA	4526
	GGCAGGTG <u>T</u> CGCGGCGA	4527
·	TCGCCGCGACACCTGCC	4528
Sulfonylurea Resistance ALS	CCGGGTTTGCTGATGCTTTGCTCGATTCCGTTCCACTGGTGGCGA TCACGGGGCAGGTGCAGGCGAATGATTGGGACGGATGCTTTT CAGGAGACTCCTATTGTTGAGGTAACACGGTC	4529
Bassia scoparia Pro189Gln CCG-CAG	GACCGTGTTACCTCAACAATAGGAGTCTCCTGAAAAGCATCCGTC CCAATCATTCGCCGCTGCACCTGCCCCGTGATCGCCACCAGTGG AACGGAATCGAGCAAAGCATCAGCAAACCCGG	4530
	GCAGGTGC <u>A</u> GCGGCGAA	4531
	TTCGCCGCTGCACCTGC	4532
Imidazolinone Resistance ALS	GACCTTACCTGCTTGATGTGATTGTACCTCATCAGGAGCATGTGC TGCCTATGATTCCTAATGGTGCAGCCTTCAAGGATATCATTAACGA AGGTGATGGAAGAACAAGTTATTGATGTTC	4533
Bassia scoparia Ser649Asn AGT-AAT	GAACATCAATAACTTGTTCTTCCATCACCTTCGTTAATGATATCCTT GAAGGCTGCACCATTAGGAATCATAGGCAGCACATGCTCCTGATG AGGTACAATCACATCAAGCAGGTAAGGTC	4534
	GATTCCTAATGGTGCAG	4535
•	CTGCACCA <u>T</u> TAGGAATC	4536
Sulfonylurea Resistance ALS 1	AGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCGC CATTACAGGACAGG	4537
Brassica napus Pro182Ser CCT-TCT	ACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTAC CGATCATCCGGCGAGAGACCTGTCCTGT	4538
·	GACAGGTC <u>T</u> CTCGCCGG	4539
	CCGGCGAGAGACCTGTC .	4540

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	Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
- [Alteration Sulfonylurea Resistance	GCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCGCC ATTACAGGACAGG	4541
	ALS 1 Brassica napus Pro182Gln	GACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTA	4542
	CCT-CAA	AACACTGTCAAGCATCGCGTCTGCTAACCCGC ACAGGTCCAACGCCGGA	4543
		TCCGGCGTTGGACCTGT	4544
	Sulfonylurea Resistance	GCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCGCC ATTACAGGACAGG	4545
	ALS 1 Brassica napus Pro182Gln	GACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTA	4546
	CCT-CAG	AACACTGTCAAGCATCGCGTCTGCTAACCCGC ACAGGTCCAGCGCCGGA	4547
		TCCGGCG <u>CT</u> GGACCTGT	4548
	Imidazolinone Resistance	GACCATACCTGTTGGATGTGATATGTCCGCACCAAGAACATGTGT TACCGATGATCCCAAATGGTGGCACTTTCAAAGATGTAATAACAGA	4549
	ALS 1 Brassica napus Ser638Asn	AGGGATGGTCGCACTAAGTACTGAGAGAT ATCTCTCAGTACTTAGTGCGACCATCCCCTTCTGTTATTACATCTT TGAAAGTGCCACCATTTGGGATCATCGGTAACACATGTTCTTGGT GCGGACATATCACATCCAACAGGTATGGTC	4550
	AGT-AAT	GCGGACATATCACATCCAACAGGTATGGTG GATCCCAAATGGTGGCA	4551
		TGCCACCATTTGGGATC	4552
	Sulfonylurea Resistance	CAGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCG CCATTACAGGACACGATCGTTGAGGTAACGAGG	4553
	ALS 2 Brassica napus Pro126Ser	CCTCGTTACCTCAACGATTGTGTCTCTTGGAAGGCGTCAGTACC GATCATCCGGCGAGGAACCTGTCCTGT	4554
	CCC-TCC	GGACAGGTTCCTCGCCG	455
		CGGCGAGG <u>A</u> ACCTGTCC	455
	Sulfonylurea Resistance	AGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCGC CATTACAGGACAGG	455
	ALS 2 Brassica napus Pro126Gln	ACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTAC CGATCATCCGGCGAGTGACCTGTCCTGT	455
	CCC-CAG	ACACTGTCAAGCATCGCGTCTGCTAACCCGCT GACAGGTCACTCGCCGG	455

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
		CCGGCGAG <u>T</u> GACCTGTC	4560
	Imidazolinone Resistance ALS 2	GACCATACCTGTTGGATGTGATATGTCCGCACCAAGAACATGTGT TACCGATGATCCCAAATGGTGGCACTTTCAAAGATGTAATAACAGA AGGGGATGGTCGCACTAAGTACTGAGAGAT	4561
5	Brassica napus Ser582Asn AGT-AAT	ATCTCTCAGTACTTAGTGCGACCATCCCCTTCTGTTATTACATCTT TGAAAGTGCCACCATTTGGGATCATCGGTAACACATGTTCTTGGT GCGGACATATCACATCCAACAGGTATGGTC	4562
		GATCCCAA <u>A</u> TGGTGGCA	4563
		TGCCACCATTTGGGATC	4564
	Sulfonylurea Resistance ALS 3	AGCGGGTTAGCCGACGCGATGCTTGACAGTGTTCCTCTCGCCCATCACAGGACAGGTCTCTCGCCGGATGATCGGTACTGACGCGTTCCAAGAGACGCCAATCGTTGAGGTAACGAGGT	4565
10	Brassica napus Pro179Ser CCT-TCT	ACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTAC CGATCATCCGGCGAGAGACCTGTCCTGT	4566
		GACAGGTC <u>T</u> CTCGCCGG	4567
		CCGGCGAG <u>A</u> GACCTGTC	4568
15	Sulfonylurea Resistance ALS 3	GCGGGTTAGCCGACGCGATGCTTGACAGTGTTCCTCTCGCC ATCACAGGACAGG	4569
	Brassica napus Pro179Gln CCT-CAA	GACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTA CCGATCATCCGGCG <u>TT</u> GGACCTGTCCTGTGATGGCGACGAGAGG AACACTGTCAAGCATCGCGTCGGCTAACCCGC	4570
		ACAGGTCC <u>AA</u> CGCCGGA	4571
		TCCGGCG <u>TT</u> GGACCTGT	4572
20	Sulfonylurea Resistance ALS 3	GCGGGTTAGCCGACGCGATGCTTGACAGTGTTCCTCTCGCC ATCACAGGACAGG	4573
	Brassica napus Pro179Gln CCT-CAG	GACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTA CCGATCATCCGGCGCTGGACCTGTCCTGT	4574
	·	ACAGGTCC <u>AG</u> CGCCGGA	4575
		TCCGGCG <u>CT</u> GGACCTGT	4576
25	Imidazolinone Resistance ALS 3	GACCGTACCTGTTGGATGTCATCTGTCCGCACCAAGAACATGTGT TACCGATGATCCCAAAATGGTGGCACTTTCAAAGATGTAATAACCG AAGGGGATGGTCGCACTAAGTACTGAGAGAT	4577
30	Brassica napus Ser635Asn AGT-AAT	ATCTCTCAGTACTTAGTGCGACCATCCCCTTCGGTTATTACATCTT TGAAAGTGCCACCATTTGGGATCATCGGTAACACATGTTCTTGGT GCGGACAGATGACATCCAACAGGTACGGTC	4578

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Alteration	GATCCCAA <u>A</u> TGGTGGCA	4579
	TGCCACCATTTGGGATC	4580
Sulfonylurea Resistance	TCCGCGCTCGCCGACGCGCTGCTCGACTCCGATGGTCG CCATCACGGGCCAGGTCTCCCGCCGCATGATCGGCACCGC CTTCCAGGAGACGCCCATAGTCGAGGTCACCCGCT	4581
ALS Oryza sativa Pro171Ser	AGCGGGTGACCTCGACTAGTCCACGTCACGTCACGTCAC	4582
CCC-TCC	GCAGGTCTCCCGCCGC	4583
	GCGGCGGAGACCTGGC	4584
Sulfonylurea Resistance	CCGCGCTCGCCGACGCGCTGCTCGACTCCGATGGTCGC CATCACGGGCCAGGTCCAACGCCGCATGATCGGCACCGACGCC	4585
ALS O <i>nyza sativa</i> Pro171Gln	TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGTTGGACCTCGCCGAGCGCGGG GCCGATCATGCACCACCGCGAGCGCGGGGGGGGGG	4586
CCC-CAA	GGACGGAGCGCGCGCGCGGGCGAGCGCGG CCAGGTCCAACGCCGCA	4587
	TGCGGCG <u>TT</u> GGACCTGG	4588
Sulfonylurea Resistance	CCGCGCTCGCCGACGCGCTGCTCGACTCCCGATGGTCGC CATCACGGGCCAGGTCCACGCCCGCTC	4589
ALS Oryza sativa Pro171Gln	TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGCTGGACCTGGCCCGTGATGGCGACCATC GGGACGAGTCGAGCAGCGCGCGAGCGCGG	4590
CCC-CAG	CCAGGTCCAGCCGCA	459
	TGCGGCG <u>CT</u> GGACCTGG	459
Imidazolinone Resistance	GGCCATACTTGTTGGATATCATCGTCCCGCACCAGGAGCATGTGC TGCCTATGATCCCAAATGGGGGCGCATTCAAGGACATGATCCTGG ATGGTGATGGCAGGACTGTGTATTAATCTAT	459
ALS Oryza sativa Ser627Asn AGT-AAT	ATAGATTAATACACAGTCCTGCCATCACCATCCAGGATCATGTCCT TGAATGCGCCCCCATTTGGGATCATAGGCAGCACATGCTCCTGGT GCGGGACGATGATATCCAACAAGTATGGCC	459
	GCGGGACGATGATATCCAACAAGTATGGGG GATCCCAAATGGGGGCG	459
	CGCCCCATTTGGGATC	459

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Sulfonylurea Resistance ALS	TCTGCGCTCGCAGACGCGTTGCTCGACTCCGTCCCCATGGTCGC CATCACGGGACAGGTG <u>T</u> CGCGACGCATGATTGGCACCGACGCCT TTCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4597
Zea mays Pro165Ser CCG-TCG	AGCGGGTGACCTCGACGATGGGCGTCTCCTGAAAGGCGTCGGTG CCAATCATGCGTCGCGACACCTGTCCCGTGATGGCGACCATGGG GACGGAGTCGAGCAACGCGTCTGCGAGCGCAGA	4598
	GACAGGTG <u>T</u> CGCGACGC	4599
·	GCGTCGCG <u>A</u> CACCTGTC	4600
Sulfonylurea Resistance ALS	CTGCGCTCGCAGACGCGTTGCTCGACTCCGTCCCCATGGTCGCC ATCACGGGACAGGTGCAGCGACGCATGATTGGCACCGACGCCTT TCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4601
Zea mays Pro165Gln CCG-CAG	GAGCGGTGACCTCGACGATGGCGTCTCCTGAAAGGCGTCGGT GCCAATCATGCGTCGCTGCACCTGTCCCGTGATGGCGACCATGG GGACGGAGTCGAGCAACGCGTCTGCGAGCGCAG	4602
	ACAGGTGC <u>A</u> GCGACGCA	4603
	TGCGTCGC <u>T</u> GCACCTGT	4604
Imidazolinone Resistance ALS	GGCCGTACCTCTTGGATATAATCGTCCCGCACCAGGAGCATGTGT TGCCTATGATCCCTAATGGTGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTATTGATCCGT	4605
Zea mays Ser621Asn AGT-AAT	ACGGATCAATACACAGTCCTGCCATCACCATCCAGGATCATATCC TTGAAAGCCCCACCATTAGGGATCATAGGCAACACATGCTCCTGG TGCGGGACGATTATATCCAAGAGGTACGGCC	4606
	GATCCCTAATGGTGGGG	4607
	CCCCACCATTAGGGATC	4608
Sulfonylurea Resistance ALS	AGTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCG ATCACTGGTCAAGTCTCTCGTCGGATGATCGGTACCGATGCTTTC CAGGAAACTCCAATTGTTGAGGTAACAAGGT	4609
Gossypium hirsutum Pro186Ser CCT-TCT	ACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTAC CGATCATCCGACGAGAGACTTGACCAGTGATCGCCACGAGAGGG ATACTATCGAGCATTGCATCAGCGAGACCACT	4610
	GTCAAGTCTC <u>T</u> CGTCGG	4611
	CCGACGAGAG <u>A</u> CTTGAC	4612
Sulfonylurea Resistance ALS	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCAACGTCGGATGATCGGTACCGATGCTTTCC AGGAAACTCCAATTGTTGAGGTAACAAGGTC	4613
Gossypium hirsutum Pro186Gln CCT-CAA	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACG <u>TT</u> GGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	4614
	TCAAGTCCAACGTCGGA	4615

Phenotype. Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Alteration	TCCGACG <u>TT</u> GGACTTGA	4616
Sulfonylurea Resistance	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCAGCGTCGGATGATCGGTACCGATGCTTTCC AGGAAACTCCAATTGTTGAGGTAACAAGGTC	4617
ALS Gossypium hirsutum Pro186Gln		4618
CCT-CAG	TCAAGTCCAGCATTCCATCATCATCATCATCATCATCATCATCATCATCA	4619
	TCCGACGCTGGACTTGA	4620
Imidazolinone Resistance	GACCTTACTTGTTGGATGTGATTGTCCCACATCAAGAACATGTCCT GCCTATGATCCCCAATGGAGGGGCTTTCAAAGATGTGATCACAGA GGGTGATGGAAGAACACAATATTGACCTCA	4621
ALS Gossypium hirsutun Ser642Asn		4622
AGT-AAT	GATCCCCAATGAGGGGG	4623
	CCCCTCCATTGGGGATC	4624
Sulfonylurea Resistance	TCTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCC ATTACTGGGCAAGTTTCCCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACTCGAT	4625
ALS Amaranthus powell Pro192Ser	ATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTACC	4626
CCC-TCC	CTGAGTCAAGAAGTGCATCAGCAAGACCAGA GGCAAGTT <u>T</u> CCCGGCGT	4627
	ACGCCGGGAAACTTGCC	4628
Sulfonylurea Resistance	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAACGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACTCGATC	4629
ALS Amaranthus powe Pro192Gln	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGTTGAACTTGCCCAGTAATGGCGACAAGAGGGA	4630
CCC-CAA	CTGAGTCAAGAAGTGCATCAGCAAGACCAG GCAAGTTCAACGGCGTA	463
	TACGCCGTTGAACTTGC	463
Sulfonylurea Resistance	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAGCGGCGTATGATTGGTACTGATGCTTTTC	463
ALS Amaranthus power Pro192Gln		463

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GCAAGTTC <u>AG</u> CGGCGTA	4635
	TACGCCG <u>CT</u> GAACTTGC	4636
Imidazolinone Resistance ALS	GACCGTATCTGCTGGATGTAATCGTACCACATCAGGAGCATGTGC TGCCTATGATCCCTAACGGTGCCGCCTTCAAGGACACCATAACAG AGGGTGATGGAAGAAGGGCTTATTAGTTGGT	4637
Amaranthus powellii Ser652Asn AGC-AAC	ACCAACTAATAAGCCCTTCTTCCATCACCCTCTGTTATGGTGTCCT TGAAGGCGGCACCGTTAGGGATCATAGGCAGCACATGCTCCTGA TGTGGTACGATTACATCCAGCAGATACGGTC	4638
	GATCCCTA <u>A</u> CGGTGCCG	4639
	CGGCACCGTTAGGGATC	4640

Table 12

<u>Genome-Altering Oligos Conferring Porphyric Herbicide Resistance</u>

	Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
	Alteration Porphyric Herbicide Resistant	TCTTGCGCCCTCTTTCTGAATCTGCTGCAAATGCACTCTCAAAACT ATATTACCCACCAATGGCAGCAGTATCTATCTCGTACCCGAAAGA AGCAATCCGAACAGAATGTTTGATAGATGG	4641
	PPO Arabidopsis thaliana Val365Met	CCATCTATCAAACATTCTGTTCGGATTGCTTCTTTCGGGTACGAGA TAGATACTGCTGCCATTGGTGGGTAATATAGTTTTGAGAGTGCATT TGCAGCAGATTCAGAAAGAGGGCGCAAGA	4642
	GTT-ATG	CCCACCAATGCAGCAG	4643
		CTGCTGC <u>C</u> A <u>T</u> TGGTGGG	4644
	Porphyric Herbicide Resistant	TATTACGTCCTCTTTCGGTTGCCGCAGCAGATGCACTTTCAAATTT CTACTATCCCCCAATGGGAGCAGTCACAATTTCATATCCTCAAGAA GCTATTCGTGATGAGCGTCTGGTTGATGG	4645
	PPO Nicotiana tabacum Val376Met	CCATCAACCAGACGCTCATCACGAATAGCTTCTTGAGGATATGAA ATTGTGACTGCTCCCATTGGGGGGATAGTAGAAATTTGAAAGTGCA TCTGCTGCGGCAACCGAAAGAGGGCGTAATA	4646
	GTT-ATG	TCCCCCAATGGGAGCAG	464
		CTGCTCC <u>C</u> A <u>T</u> TGGGGGA	464
	Porphyric Herbicide Resistant	TGTTGCGTCCGCTTTCGTTGGGTGCAGCAGATGCATTGTCAAAAT TTTATTATCCTCCGATGGCAGCTGTATCAATTTCATATCCAAAAGA CGCAATTCGTGCTGACCGGCTGATTGATGG	464
	PPO Cichorium intybus Val383Met	CCATCAATCAGCCGGTCAGCACGAATTGCGTCTTTTGGATATGAA ATTGATACAGCTGCCATCGGAGGATAATAAAATTTTGACAATGCAT CTGCTGCACCCAACGAAAGCGGACGCAACA	465
	GTT-ATG	TCCTCCGATGCAGCTG	465
		CAGCTGC <u>C</u> A <u>T</u> CGGAGGA	465
	Porphyric Herbicide Resistant PPO Spinacia oleracea Val390Met	TCCTTCGTCCACTTTCAGATGTCGCCGCAGAATCTCTTTCAAAATT TCATTATCCACCAATGGCAGCTGTGTCACTTTCCTATCCTAAAGAA GCAATTAGATCAGAGTGCTTGATTGACGG	46
		CCGTCAATCAGACCACTCGATCTAATTGCTTCTTTAGGATAGGAAA GTGACACAGCTGCCATTGGTGGATAATGAAATTTTGAAAGAGATTC TGCGGCGACATCTGAAAGTGGACGAAGGA	<u> </u>
	GTT-ATG	TCCACCAATGGCAGCTG	46
		CAGCTGCCATTGGTGGA	46

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Porphyric Herbicide Resistant PPO	TTTTGCGTCCACTTTCAAGCGATGCTGCAGATGCTCTATCAAGATT CTATTATCCACCGATGGCTGCTGTAACTGTTTCGTATCCAAAGGAA GCAATTAGAAAAGAATGCTTAATTGATGG	4657
Zea mays Val363Met GTT-ATG	CCATCAATTAAGCATTCTTTTCTAATTGCTTCCTTTGGATACGAAAC AGTTACAGCAGCCATCGGTGGATAATAGAATCTTGATAGAGCATC TGCAGCATCGCTTGAAAGTGGACGCAAAA	4658
	TCCACCGATGGCTGCTG	4659
	CAGCAGC <u>C</u> A <u>T</u> CGGTGGA	4660
Porphyric Herbicide Resistant PPO	TCTTGCGGCCACTTTCAAGTGATGCAGCAGATGCTCTGTCAATATT CTATTATCCACCAATGGCTGCTGTAACTGTTTCATATCCAAAAGAA GCAATTAGAAAAGAATGCTTAATTGACGG	4661
Oryza sativa Val364Met GTT-ATG	CCGTCAATTAAGCATTCTTTTCTAATTGCTTCTTTTGGATATGAAAC AGTTACAGCAGCCATTGGTGGATAATAGAATATTGACAGAGCATCT GCTGCATCACTTGAAAGTGGCCGCAAGA	4662
	TCCACCAATGGCTGCTG	4663
	CAGCAGC C A <u>T</u> TGGTGGA	4664
Porphyric Herbicide Resistant PPO	CTGGTCAAGGAGCAGGCGCCGCCGCCGCCGAGGCCCTGGGCT CCTTCGACTACCCGCCGATGGGCGCCGTGACGCTGTCGTACCCG CTGAGCGCCGTGCGGGAGGAGCGCAAGGCCTCGG	4665
Chlamydomonas reinhardtii Val389Met	CCGAGGCCTTGCGCTCCTCCCGCACGGCGCTCAGCGGTACGA CAGCGTCACGGCGCCCATCGGCGGGTAGTCGAAGGAGCCCAGG GCCTCGGCGGCGGCGGCGCCTGCTCCTTGACCAG	4666
GTG-ATG	ACCCGCCG <u>A</u> TGGGCGCC	4667
	GGCGCCCA <u>T</u> CGGCGGGT	4668

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Table 13
Genome-Altering Oligos Conferring Triazine Resistance

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Triazine Resistant	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT	4669
D1 Protein Arabidopsis thaliana	TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	
Ser264Thr	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA	4670
AGT-ACT	CGAGAATTGTTGAAA <u>G</u> TAGCATATTGGAAAATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	ATATGCTA <u>C</u> TTTCAACA	4671
•	TGTTGAAAGTAGCATAT	4672
Triazine Resistant D1 Protein <i>Nicotiana tabacum</i> Ser264Thr AGT-ACT	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4673
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4674
	ATATGCTACTTTCAACA	4675
	TGTTGAAAGTAGCATAT	4676
Triazine Resistant D1 Protein Populus deltoides	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACCTTTTAACAACTCTCGCTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATCTG	4677
Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAG CGAGAGTTGTTAAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCGGCTACGATATTATAAGTTT	4678
	ATATGCTACTTTTAACA	4679
	TGTTAAAAGTAGCATAT	4680
Triazine Resistant D1 Protein	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4681
Petunia x hybrida Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATATTATAAGTTT	4682
	ATATGCTACTTTCAACA	4683
	TGTTGAAA G TAGCATAT	4684
Triazine Resistant D1 Protein Magnolia pyramidata Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4685
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACGATATTATAAGTTT	4686
	ATATGCTA <u>C</u> TTTCAACA	4687
	TGTTGAAAGTAGCATAT	4688
Triazine Resistant D1 Protein Medicago sativa	AAACCTATAATATTGTAGCAGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4689
Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCTGCTACAATATTATAGGTTT	4690
	ATATGCTA <u>C</u> TTTCAACA	4691

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGTTGAAA G TAGCATAT	4692
Triazine Resistant D1 Protein Glycine max Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCAACCTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4693
	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTTGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACAATATTATAGGTTT	4694
	ATATGCAACA	4695
	TGTTGAAA G TTGCATAT	4696
Triazine Resistant D1 Protein Brassica napus Gly264Thr GGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4697
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4698
	ATATGCT <u>AC</u> TTTCAACA	4699
	TGTTGAAA GT AGCATAT	4700
Triazine Resistant D1 Protein Oryza sativa Ser264Thr AGT-ACT	AAACTTATAATATTGTGGCCGCTCATGGTTATTTTGGCCGATTAAT CTTCCAATATGCTACTTTTAACAACTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATTTG	4701
	CAAATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTAAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCGGCCACAATATTATAAGTTT	4702
	ATATGCTACTTTTAACA	4703
	TGTTAAAA G TAGCATAT	4704
Triazine Resistant D1 Protein Zea mays	AGACTTATAATATTGTGGCTGCTCACGGTTATTTTGGTCGATTAAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATCTG	4705
Ser264Thr AGT-ACT	CAGATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATTAATCGACCAAAAT AACCGTGAGCAGCCACAATATTATAAGTCT	4706
	ATATGCTA <u>C</u> TTTCAACA	4707
	TGTTGAAA G TAGCATAT	4708
Triazine Resistant D1 Protein Arabidopsis thaliana Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	4709
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAAATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4710
	ATATGCTA <u>C</u> TTTCAACA	4711
	TGTTGAAAGTAGCATAT	4712

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT	4713
riazine Resistant	CTTCCAATATGCTACTTCAACAACTCTCGTTCGTTACACTTCTTCC	
)1 Protein	TAGCTGCTTGGCCTGTAGTAGGTATCTG	
licotiana tabacum	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA	4714
Ser264Thr	CGAGAGTTGTTGAAA <u>G</u> TAGCATATTGGAAGATCAATCGGCCAAAA	
AGT-ACT	TAACCATGAGCGGCTACGATGTTATAAGTTT	_
	ATATGCTACTTTCAACA	4715
		4716
	TGTTGAAAGTAGCATAT	4717
Triazine Resistant	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT	7/1/
D1 Protein	CTTCCAATATGCTACTTTTAACAACTCTCGCTCTTTACATTTCTTCT	
Populus deltoides	TAGCTGCTTGGCCTGTAGTAGGTATCTG	4718
Ser264Thr	CAGATACCTACTACAGGCCAAGCATCAATCGCCCAAAAT	47.10
AGT-ACT	CGAGAGTTGTTAAAAGTTAGCATATTATAACTTT	
	AACCATGAGCGGCTACGATATTATAAGTTT	4719
	ATATGCTA <u>C</u> TTTTAACA	4720
	TGTTAAAAGTAGCATAT	4721
Triazine Resistant	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT	4/2
D1 Protein	CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC	
Petunia x hybrida	TAGCTGCTTGGCCTGTAGTAGGTATCTG	4722
Ser264Thr	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA	4122
AGT-ACT	CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA	
	TAACCATGAGCGGCTACGATATTATAAGTTT	472
	ATATGCTA <u>C</u> TTTCAACA	472
	TGTTGAAAGTAGCATAT	
Triazine Resistant	AAACTTATAATATCGTAGCTGCTCATGGTTATTTTGGCCGATTGAT	472
D1 Protein	CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCC	1
Magnolia pyramidata	TAGCTGCTTGGCCTGTAGTAGGTATCTG	472
Ser264Thr	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA	i i
AGT-ACT	CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT	1
	AACCATGAGCAGCTACGATATTATAAGTTT	472
	ATATGCTA <u>C</u> TTTCAACA	
	TGTTGAAA G TAGCATAT	472
Triazine Resistant	AAACCTATAATATTGTAGCAGCTCATGGTTATTTTGGCCGATTGAT	472
D1 Protein	CTTCCAATATGCTACTTTCAACAACTCTCGTTCTTTACATTCTTCC	
Medicago sativa	TACCTCCTTCGCCTGTAGTAGGTATCTG	
Ser264Thr	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA	4/3
AGT-ACT	CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA	
	TAACCATGAGCTGCTACAATATTATAGGTTT	473
	ATATGCTACTTTCAACA	
	TGTTGAAAGTAGCATAT	47

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Triazine Resistant D1 Protein Glycine max Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCAACACAATTCTCGTTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4733
	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAA G TTGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACAATATTATAGGTTT	4734
	ATATGCAACA .	4735
	TGTTGAAA <u>G</u> TTGCATAT	4736
Triazine Resistant D1 Protein Brassica napus	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCT <u>AC</u> TTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4737
Gly264Thr GGT-ACT	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4738
	ATATGCT <u>AC</u> TTTCAACA	4739
	TGTTGAAA <u>GT</u> AGCATAT	4740
Triazine Resistant D1 Protein Onyza sativa Ser264Thr AGT-ACT	AAACTTATAATATTGTGGCCGCTCATGGTTATTTTGGCCGATTAAT CTTCCAATATGCTACTTTTAACAACTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATTTG	4741
	CAAATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTAAAA <u>GT</u> AGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCGGCCACAATATTATAAGTTT	4742
	ATATGCTACTTTTAACA	4743
	TGTTAAAA G TAGCATAT	4744
Triazine Resistant D1 Protein Zea mays Ser264Thr AGT-ACT	AGACTTATAATATTGTGGCTGCTCACGGTTATTTTGGTCGATTAAT CTTCCAATATGCTACTTCAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATCTG	4745
	CAGATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGAACCGAGAATTGTTGAAAGTAGCATATTGGAAGATTAATCGACCAAAATAACCGTGAGCAGCCACAATATTATAAGTCT	4746
	ATATGCTA <u>C</u> TTTCAACA	4747
	TGTTGAAAGTAGCATAT	4748
Triazine Resistant D1 Protein Arabidopsis thaliana Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	4749
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAAATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4750
	ATATGCTA <u>C</u> TTTCAACA	4751
	TGTTGAAAGTAGCATAT	4752

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	AAACCTACAATATTGTGGCTGCTCACGGTTATTTCGGCCGATTGAT	4753
riazine Resistant	CTTCCAGTATGCTACTTTCAACAACTCCCGTTCTTTACATTTCTTCT	
01 Protein	TAGCTGCTTGGCCCGTAGCAGCAGCTATCTG TAGCTGCTTGGCCCGTAGCAGCAGCTATCTG	
Picea abies	CAGATACCTGCTACGGGCCAAGCAGCTAAGAAGAATGTAAAGAA	4754
Ser264Thr	CGGGAGTTGTTGAAAGTAGCATACTGGAAGATCAATCGGCCGAAA	
AGT-ACT	TAACCGTGAGCAGCACAATATTGTAGGTTT	
	GTATGCTACTTTCAACA	4755
		4756
	TGTTGAAAGTAGCATAC	4757
Triazine Resistant	AAACCTATAATATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT	4707
D1 Protein	CTTCCAATATGCTACTTTCAACAATTCTCGCTCTTTACATTTCTCC	
Vicia faba	TAGCTGCTTGGCCTGTAGTAGGTATCTG CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAATGTAAAGAG	4758
Ser264Thr	CAGATACCTACTACAGGCCAAGCTAGGAAGAACTCTTTTTTTT	
AGT-ACT	CGAGAATGTGAAAGTAGCATATTATAGGTT	
	AACCGTGAGCAGCTACAATATTATAGGTTT	4759
	ATATGCTA <u>C</u> TTTCAACA	4760
	TGTTGAAAGTAGCATAAT	4761
Triazine Resistant	AGACTTATAATATTGTGGCTGCTCATGGTTATTTTGGCCGATTAAT	4/01
D1 Protein	CTTCCAATATGCTACTTTCAACAACTCTCGTTCTTTACACTTCTTCT	
Hordeum vulgare	TGGCTGCTTGGCCTGTAGTAGGAACTCTAAAGAA	4762
Ser264Thr	CAGATTCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA	4102
AGT-ACT	CGAGAGTTGTTGAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT	
	AACCATGAGCAGCACAATATTATAAGTCT	4763
	ATATGCTACTTCAACA	4764
	TGTTGAAAGTAGCATAT	
Triazine Resistant	AAACTTATAATATTGTGGCTGCTCATGGTTATTTTGGCCGATTAAT	476
D1 Protein	CTTCCAATATGCTACTTTCAACAACTCTCGTTCTTTACACTTCTTCT	
Triticum aestivum	TGGCTGCTTGGCCTGTAGTAGGAATCTG	476
Ser264Thr	CAGATTCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA	4/0
AGT-ACT	CGAGAGTTGTTGAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT	
	AACCATGAGCAGCCACAATATTATAAGTTT	476
	ATATGCTACTTTCAACA	476
	TGTTGAAA G TAGCATAT	
Triazine Resistant	AAACTTATAATATTGTAGCTGCTCATGGTTATTTTGGCCGATTAATC	476
D1 Protein	TTCCAATATGCAACTTTCAACAATTCTCGTTCTTTACATTTCTTCCT	1
Vigna unguiculata	AGCTGCTTGGCCTGTAGTAGGTATTTG	1-7-
Ser264Thr	CAAATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA	477
AGT-ACT	CGAGAATTGTTGAAAGTTGCATATTGGAAGATTAATCGGCCAAAAT	
	AACCATGAGCAGCTACAATATTATAAGTTT	477
	ATATGCAACATTTCAACA	
1	TGTTGAAAGTTGCATAT	477

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Triazine Resistant D1 Protein Lotus japonicus	AAACCTATAATATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCAACTTTCAACAACTCTCGTTCTTTACACTTCTTCT TAGCTGCTTGGCCTGTTGTAGGTATCTG	4773
Ser264Thr AGT-ACT	CAGATACCTACAACAGGCCAAGCAGCTAAGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTTGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATATTATAGGTTT	4774
	ATATGCAACA TITCAACA	4775
	TGTTGAAA <u>G</u> TTGCATAT	4776
Triazine Resistant D1 Protein Sinapis alba	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4777
Ser264Thr AGT-ACT	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAA G TAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4778
	ATATGCTA <u>C</u> TTTCAACA	4779
	TGTTGAAA <u>G</u> TAGCATAT	4780
Triazine Resistant D1 Protein Pisum sativum	AAACCTATAATATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGCTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4781
Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAATGTAAAGAG CGAGAATTGTTGAAA <u>G</u> TAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATATTATAGGTTT	4782
	ATATGCTA <u>C</u> TTTCAACA	4783
	TGTTGAAA <u>G</u> TAGCATAT	4784
Triazine Resistant D1 Protein Spinacia oleracea	AAACTTATAATATCGTAGCTGCTCATGGTTATTTTGGTCGATTGAT CTTCCAATATGCTACCTTCAACAACTCTCGTTCTTTACACTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4785
Ser264Thr AGT-ACT	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGACCAAAAT AACCATGAGCAGCTACGATATTATAAGTTT	4786
	ATATGCTA <u>C</u> TTTCAACA	4787
	TGTTGAAA <u>G</u> TAGCATAT	4788
Triazine Resistant D1 Protein Nicotiana debneyi	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4789
Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4790
	ATATGCTA <u>C</u> TTTCAACA	4791
	TGTTGAAAGTAGCATAT	4792

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Triazine Resistant D1 Protein	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4793
Solanum nigrum Ser264Thr AGT-ACT	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATATTATAAGTTT	4794
	ATATGCTACTTTCAACA	4795
	TGTTGAAA G TAGCATAT	4796
Triazine Resistant D1 Protein	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4797
Nicotiana plumbaginifolia Ser264Thr	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4798
AGT-ACT	ATATGCTACTTCAACA	4799
	TGTTGAAA <u>G</u> TAGCATAT	4800

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Example 6 Engineering male- or female-sterile plants

Flower development in distantly related dicot plant species is increasingly better understood and appears to be regulated by a family of genes which encode regulatory proteins. These genes include, for example, AGAMOUS (AG), APETALA1 (AP1), and APETALA3 (AP3) and PISTILLATA (PI) in Arabidopsis thaliana, and DEFICIENS A (DEFA), GLOBOSA (GLO), SQUAMOSA (SQUA), and PLENA (PLE) in Antirrhinum majus. Genetic studies have shown that the DEFA, GLO and AP3 genes are essential for petal and stamen development. Sequence analysis of these genes revealed that the gene products contain a conserved MADS box region, a DNA-binding domain. Using these clones as probes, MADS box genes have also been isolated from other species including tomato, tobacco, petunia, Brassica napus, and maize.

Altering the expression of these genes results in altered floral morphology. For example, mutations in *AP3* and *PI* result in male-sterile flowers because petals develop in place of stamens.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that confer altered floral structures in plants.

Table 14
Oligonucleotides to produce male-sterile plants

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile	TTGTCCTCTCCACCAAATCTCTTCAACAAAAAGATTAAACAAAGAGA	4801
AP3 Arabidopsis thaliana	GAAGAATATGGCG <u>T</u> GAGGGAAGATCCAGATCAAGAGGATAGAGAA CCAGACAAACAGACAAGTGACGTATTCAA	
Arg3Term AGA-TGA	TTGAATACGTCACTTGTCTGTTTGTCTGGTTCTCTATCCTCTTGATC TGGATCTTCCCTCACGCCATATTCTTCTCTCTTTTGTTTAATCTTTTT GTTGAAGAGATTTGGTGGAGAGACAA	4802
	ATATGGCG <u>T</u> GAGGGAAG	4803
	CTTCCCTC <u>A</u> CGCCATAT	4804
Male-sterile AP3 Arabidopsis thaliana	TCTCCACCAAATCTCTTCAACAAAAGATTAAACAAAGAGAGAG	4805
Lys5Term AAG-TAG	TTCTCTTTGAATACGTCACTTGTCTGTTTGTCTGGTTCTCTATCCTC TTGATCTGGATCTACCCTCTCGCCATATTCTTCTCTCTTTGTTTAAT CTTTTTGTTGAAGAGATTTGGTGGAGA	4806
	CGAGAGGG <u>T</u> AGATCCAG	4807

	CTGGATCTACCCTCTCG	4808
1ale-sterile NP3	CCAAATCTCTTCAACAAAAGATTAAACAAAGAGAGAAGAATATGG CGAGAGGGAAGATCTAGATCAAGAGGATAGAGAACCAGACAAACA	4809
Arabidopsis thaliana GIn7Term CAG-TAG	GACAAGTGACGTATTCAAAGAGAAGGAATG CATTCCTTCTCTTTGAATACGTCACTTGTCTGTTTGTCTGGTTCTCT ATCCTCTTGATCTAGATCTTCCCTCTCGCCATATTCTTCTCTCTTTG TTTAATCTTTTTGTTGAAGAGATTTGG	4810
	GGAAGATCTAGATCAAG	4811
	CTTGATCT <u>A</u> GATCTTCC	4812
Male-sterile AP3	CTCTTCAACAAAAGATTAAACAAAGAGAGAAGAATATGGCGAGAG GGAAGATCCAGATC <u>T</u> AGAGGATAGAGAACCAGACAAACAGACAAG TGACGTATTCAAAGAGAAGGAATGGTTTAT	4813
Arabidopsis thaliana Lys9Term AAG-TAG	ATAAACCATTCCTTCTCTTTGAATACGTCACTTGTCTGTTTGTCTGG TTCTCTATCCTCTAGATCTGGATCTTCCCTCTCGCCATATTCTTCTC	4814
	TCTTTGTTTAATCTTTTTGTTGAAGAG TCCAGATCTAGAGGATA	4815
	TATCCTCTAGATCTGGA	4816
Male-sterile AP3	AGAGGGAAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGA CAAGTGACGTATTCTTAGAGAAGAAATGGTTTGTTCAAGAAAGCTC ACGAGCTTACAGTTTTATGTGATGCTAGGG	4817
Brassica oleracea Lys23Term AAG-TAG	CCCTAGCATCACATATATGTGATCOTAGGGGGGGGGGGGG	4818
	CGTATTCT <u>T</u> AGAGAAGA	4819
	TCTTCTCT <u>A</u> AGAATACG	4820
Male-sterile AP3	GGGAAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGACAA GTGACGTATTCTAAGTGAAGAAATGGTTTGTTCAAGAAAGCTCACG AGCTTACAGTTTTATGTGATGCTAGGGTTT	482
Brassica oleracea Arg24Term AGA-TGA	AAACCTAGCATCACATAAAACTGTAAGCTCGTGAGCTTTCTTGAA CAAACCATTTCTTCACTTAGAATACGTCACTTGTCGGTTGGTCTGG TTCTCTATCCTCTTGATCTGGATCTTCCC	482
	ATTCTAAG <u>T</u> GAAGAAAT	482
	ATTTCTTCACTTAGAAT	482
Male-sterile AP3 Brassica oleracea	AAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGACAAGTG ACGTATTCTAAGAGA <u>T</u> GAAATGGTTTGTTCAAGAAAGCTCACGAGC TTACAGTTTTATGTGATGCTAGGGTTTCGA	
Arg25Term AGA-TGA	TCGAAACCCTAGCATCACATAAAACTGTAAGCTCGTGAGCTTTCTT GAACAAACCATTTCATCTCTTAGAATACGTCACTTGTCGGTTGGTC TGGTTCTCTATCCTCTTGATCTGGATCTT	
	CTAAGAGA <u>T</u> GAAATGGT	482
	ACCATTTCATCTCTTAG	482

	Male-sterile AP3 Brassica oleracea	TCAAGAGGATAGAGAACCAGACCAACCGACAAGTGACGTATTCTA AGAGAAGAAATGGTTAGTTCAAGAAAGCTCACGAGCTTACAGTTTT ATGTGATGCTAGGGTTTCGATTATCATGTT	4829
5	Leu28Term TTG-TAG	AACATGATAATCGAAACCCTAGCATCACATAAAACTGTAAGCTCGT GAGCTTTCTTGAACTAACCATTTCTTCTCTTAGAATACGTCACTTGT CGGTTGGTCTGGTTCTCTATCCTCTTGA	4830
		AAATGGTT <u>A</u> GTTCAAGA	4831
		TCTTGAAC <u>T</u> AACCATTT	4832
	Male-sterile AP3 <i>Brassica napus</i>	GGCTCGAGGGAAGATCCAGATTAAGAGGATAGAGAACCAAACAAA	4833
10	Tyr21Term TAC-TAG	AGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAACAAACC ATTTCTTCTCTTGGACTAGGTGACCTGCCTGTTTGTTTGGTTCTCTA TCCTCTTAATCTGGATCTTCCCTCGAGCC	4834
		GTCACCTA <u>G</u> TCCAAGAG	4835
		CTCTTGGA <u>C</u> TAGGTGAC	4836
•	Male-sterile AP3 <i>Brassica napus</i>	CGAGGGAAGATCCAGATTAAGAGGATAGAGAACCAAACAACAGG CAGGTCACCTACTCCTAGAGAAGAAATGGTTTGTTCAAGAAAGCAC ACGAGCTCTCTGTTCTCTGTGATGCTAAAG	4837
15	Lys23Term AAG-TAG	CTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAACAA ACCATTTCTTCTCTAGGAGTAGGTGACCTGCCTGTTTGTT	4838
	-	CCTACTCC <u>T</u> AGAGAAGA	4839
		TCTTCTCT <u>A</u> GGAGTAGG	4840
	Male-sterile AP3 Brassica napus	GGGAAGATCCAGATTAAGAGGATAGAGAACCAAACAACAGGCAG GTCACCTACTCCAAG <u>T</u> GAAGAAATGGTTTGTTCAAGAAAGCACACG AGCTCTCTGTTCTCTGTGATGCTAAAGTTT	4841
20	Arg24Term AGA-TGA	AAACTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAA CAAACCATTTCTTCACTTGGAGTAGGTGACCTGCCTGTTTGTT	4842
		ACTCCAAG <u>T</u> GAAGAAAT	4843
		ATTTCTTC A CTTGGAGT	4844
	Male-sterile AP3 Brassica napus	AAGATCCAGATTAAGAGGATAGAGAACCAAACAAACAGGCAGG	4845
25	Arg25Term AGA-TGA	TGGAAACTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTT GAACAAACCATTTCATCTTTGGAGTAGGTGACCTGCCTGTTTGTT	4846
		CCAAGAGA <u>T</u> GAAATGGT	4847
		ACCATITC <u>A</u> TCTCTTGG	4848

	Male-sterile DEFA	AGTGGTTCGATGGCTTGAGGGAAGATCCAGATTAAGAGGATAGAG	4849
5	Antirrhinum majus Arg3Term CGA-TGA	AACCAAACAACAGGCAGGTCACCTACTCCA TGGAGTAGGTGACCTGCCTGTTTGTTTGGTTCTCTATCCTCTTAAT CTGGATCTTCCCTCAAGCCATCGAACCACTACCACTACTGCTCTTG	4850
		TTTTCTTCCAGCTTTCCTTTCTCCC CGATGGCTTGAGGGAAG	4851
		CTTCCCTCAAGCCATCG	4852
	Male-sterile DEFA	AAAGGAAAGCTGGAAGAAGAAAACAAGAGCAGTAGTGGTAGTGGT TCGATGGCTCGAGGGTAGATCCAGATTAAGAGGATAGAGAACCAA ACAAACAGGCAGGTCACCTACTCCAAGAGAA	4853
10	Antirrhinum majus Lys5Term AAG-TAG	TTCTCTTGGAGTAGCTGACCTGCTGTTTGTTTGGTTCTCTATCCT CTTAATCTGGATCTACCCTCGAGCCATCGAACCACTACCACTACTG CTCTTGTTTTCTTCCTGCAGCTTTCCTTT	4854
		CTCGAGGGTAGATCCAG	4855
		CTGGATCT <u>A</u> CCCTCGAG	4856
	Male-sterile DEFA	AAGCTGGAAGAAAAACAAGAGCAGTAGTGGTAGTGGTTCGATG GCTCGAGGGAAGATCTAGATTAAGAGGATAGAAACCAAACAAA	4857
15	Antirrhinum majus Gln7Term CAG-TAG	CATTTCTTCTTGGAGTAGGTGACCTGCCTGTTTGTTTGGTTCTC TATCCTCTTAATCTAGATCTTCCCTCGAGCCATCGAACCACTACCA CTACTGCTCTTGTTTTCTTCCTCCAGCTT	4858
		GGAAGATC <u>T</u> AGATTAAG	4859
		CTTAATCTAGATCTTCC	4860
	Male-sterile DEFA	GAAGAAGAAACAAGAGCAGTAGTGGTAGTGGTTCGATGGCTCGA GGGAAGATCCAGATT <u>T</u> AGAGGATAGAGAACCAAACAAACAGGCAG GTCACCTACTCCAAGAGAAGAAATGGTTTGT	4861
20	Antirrhinum majus Lys9Term AAG-TAG	ACAAACCATTTCTTCTCTTGGAGTAGGTGACCTGCCTGTTTGTT	4862
		TCCAGATT <u>T</u> AGAGGATA	4863
		TATCCTCTAAATCTGGA	4864
	Male-sterile AP3	TCAGTAATTCTTAAGATCTCAAACTTTGAGCAAAAAGAAAAAAAA	4865
25	Nicotiana tabacum Lys5Term AAG-TAG	TTCTCTTAGAATAAGTGACTTGTCTGTTTGTTTGGTTCTCTATTCTC TTGATCTGGATCTACCCCACGAGCCATAGTTTTTTTTTT	4866
		AAAGTTTGAGATCTTAAGAATTACTGA CTCGTGGGTAGATCCAG	4867
		CTGGATCTACCCACGAG	4868

Male-sterile AP3 Nicotiana tabacum	ATTCTTAAGATCTCAAACTTTGAGCAAAAAGAAAAAAAAA	486
GIn7Term CAG-TAG	CATTTCTTCTAGAATAAGTGACTTGTTTGTTTGGTTCTCT ATTCTCTTGATCTAGAATAAGTGACTTGTCTGTTTGTTTG	48
	GGAAGATC <u>T</u> AGATCAAG	48
	CTTGATCT A GATCTTCC	48
Male-sterile AP3 Nicotiana tabacum	AAGATCTCAAACTTTGAGCAAAAAGAAAAAAAAAAACTATGGCTCGTG GGAAGATCCAGATC <u>T</u> AGAGAATAGAGAACCAAACAAACAGACAAGT CACTTATTCTAAGAGAAAGAAATGGACTTT	48
Lys9Term AAG-TAG	AAAGTCCATTTCTTCTCTTAGAATAAGTGACTTGTCTGTTTGGTTTGG TTCTCTATTCTCTAGATCTGGATCTTCCCACGAGCCATAGTTTTTTT TTCTTTTTGCTCAAAGTTTGAGATCTT	48
·	TCCAGATC <u>T</u> AGAGAATA	48
	TATTCTCT A GATCTGGA	48
Male-sterile AP3 Nicotiana tabacum	ATCTCAAACTTTGAGCAAAAAGAAAAAAAAAAACTATGGCTCGTGGGA AGATCCAGATCAAGTGAATAGAGAACCAAACAAACAGACAAGTCAC TTATTCTAAGAGAAGAAATGGACTTTTCA	48
Arg10Term AGA-TGA	TGAAAAGTCCATTTCTTCTCTTAGAATAAGTGACTTGTCTGTTTGTT	48
	AGATCAAG <u>T</u> GAATAGAG	48
	CTCTATTC <u>A</u> CTTGATCT	48
Male-sterile AP3 Medicago sativa	GGCTCGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAA CAGACAAGTAACTTAGTCAAAACGAAGGGATGGTCTTTTCAAGAAG GCCAATGAGCTCACTGTTCTTTGTGATGCT	48
Tyr21Term TAC-TAG	AGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAGACCA TCCCTTCGTTTTGACTAAGTTACTTGTCTGTTCGTTGTGTTCTCTAT TCTCTTGATCTGGATCTTTCCTCGAGCC	48
	GTAACTTA G TCAAAACG	48
	CGTTTTGA <u>C</u> TAAGTTAC	48
Male-sterile AP3 Medicago sativa	CTCGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACA GACAAGTAACTTACT G AAAACGAAGGGATGGTCTTTTCAAGAAGGC CAATGAGCTCACTGTTCTTTGTGATGCTAA	48
Ser22Term TCA-TGA	TTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAGAC CATCCCTTCGTTTTCAGTAAGTTACTTGTCTGTTCGTTGTGTTCTCT ATTCTCTTGATCTGGATCTTTCCTCGAG	48
	AACTTACT G AAAACGAA	48
	TTCGTTTTCAGTAAGTT	48

	Male-sterile AP3	CGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACAGA CAAGTAACTTACTCATAACGAAGGGATGGTCTTTTCAAGAAGGCCA	4889
5	Medicago sativa Lys23Term AAA-TAA	ATGAGCTCACTGTTCTTTGTGATGCTAAGG CCTTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAG ACCATCCCTTCGTTATGAGTAAGTTACTTGTCTGTTCTTCTTCTTCTTCTTCTTCTTCTTCTTCTT	4890
		CTATTCTCTTGATCTGGATCTTTCCTCG CTTACTCATAACGAAGG	4891
		CCTTCGTT <u>A</u> TGAGTAAG	4892
	Male-sterile AP3	GGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACAGACAA GTAACTTACTCAAAATGAAGGGATGGTCTTTTCAAGAAGGCCAATG AGCTCACTGTTCTTTGTGATGCTAAGGTTT	4893
10	Medicago sativa Arg24Term CGA-TGA	AAACCTTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAA AAGACCATCCCTTCATTTTGAGTAAGTTACTTGTCTGTTCGTTGTGT TCTCTATTCTCTTGATCTGGATCTTTCC	4894
	1	ACTCAAAA <u>T</u> GAAGGGAT	4895
		ATCCCTTCATTTTGAGT	4896
	Male-sterile DEF4	GGCTCGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAA	4897
15	Solanum tuberosum Tyr21Term TAT-TAG	GCTAATGAACTTACAGTTCTTTGTGATGCT AGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGCCCA TTTCTTCTCTTTGACTAAGTCACTACCACCC	4898
		TTCTTGATCTGGATCTTACCACGAGCC GTGACTTAGTCAAAGAG	4899
		CTCTTTGACTAAGTCAC	4900
	Male-sterile DEF4	CTCGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAA	4901
20	Solanum tuberosum Ser22Term TCA-TGA	AATGAACITACAGTTCTTTGTGATGCTAA TTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGCC CATTTCTTCTCTTTCAATAAGTCACTTGCCTATTTGTTTG	4902
		GACTTATT GAAAGAGAA	4903
		TTCTCTTTCAATAAGTC	4904
	Male-sterile DEF4	CGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAA	
25	Solanum tuberosum Lys23Term AAG-TAG	CTTTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGC CCATTTCTTCTCTATGAATAAGTCACTTGCCTATTTGTTTG	
		CTTATTCATAGAGAAGA	4907
		TCTTCTCTATGAATAAG	4908

	Male-sterile DEF4	GGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAATAGGCAAG TGACTTATTCAAAG <u>T</u> GAAGAAATGGGCTATTCAAGAAGGCTAATGA	4909
5	Solanum tuberosum Arg24Term AGA-TGA	ACTTACAGTTCTTTGTGATGCTAAAGTTT AAACTTTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAAT AGCCCATTTCTTCACTTTGAATAAGTCACTTGCCTATTTGTTTG	4910
		TTCTATTTTCTTGATCTGGATCTTACC ATTCAAAG T GAAGAAAT	4911
		ATTTCTTC <u>A</u> CTTTGAAT	4912
	Male-sterile AP3 Lycopersicon	GCTAATGAACTTACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTAT GATTTCTAGTACTTGAAAACTTCATGAGTTTATAAGTCCCTCTATCA CGACCAAACAATTGTTCGATCTGTACC	4913
10	esculentum Gly27Term GGA-TGA	GGTACAGATCGAACAATTGTTTGGTCGTGATAGAGGGACTTATAAA CTCATGAAGTTTTCAAGTACTAGAAATCATAACAATTGAAACTTTAG CATCACAAAGAACAGTAAGTTCATTAGC	4914
		CTAGTACT <u>T</u> GAAAACTT	4915
		AAGTTTTC <u>A</u> AGTACTAG	4916
	Male-sterile AP3 Lycopersicon	AATGAACTTACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTATGAT TTCTAGTACTGGATAACTTCATGAGTTTATAAGTCCCTCTATCACGA CCAAACAATTGTTCGATCTGTACCAGA	4917
15	esculentum Lys28Term AAA-TAA	TCTGGTACAGATCGAACAATTGTTTGGTCGTGATAGAGGGACTTAT AAACTCATGAAGTTATCCAGTACTAGAAATCATAACAATTGAAACTT TAGCATCACAAAGAACAGTAAGTTCATT	4918
		GTACTGGA <u>T</u> AACTTCAT	4919
	}	ATGAAGTT <u>A</u> TCCAGTAC	4920
20	Male-sterile AP3 Lycopersicon	ACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTATGATTTCTAGTAC TGGAAAACTTCATTAGTTTATAAGTCCCTCTATCACGACCAAACAAT TGTTCGATCTGTACCAGAAGACTATTG	4921
	esculentum Glu31Term GAG-TAG	CAATAGTCTTCTGGTACAGATCGAACAATTGTTTGGTCGTGATAGA GGGACTTATAAACT <u>A</u> ATGAAGTTTTCCAGTACTAGAAATCATAACAA TTGAAACTTTAGCATCACAAAGAACAGT	4922
		AACTTCAT <u>T</u> AGTTTATA	4923
		TATAAACT <u>A</u> ATGAAGTT	4924
25	Male-sterile AP3 Lycopersicon	ATTGTTATGATTTCTAGTACTGGAAAACTTCATGAGTTTATAAGTCC CTCTATCACGACC <u>T</u> AACAATTGTTCGATCTGTACCAGAAGACTATT GGAGTTGATATTTGGACTACTCACTATG	4925
	esculentum Lys40Term AAA-TAA	CATAGTGAGTAGTCCAAATATCAACTCCAATAGTCTTCTGGTACAG ATCGAACAATTGTT A GGTCGTGATAGAGGGACTTATAAACTCATGA AGTTTTCCAGTACTAGAAATCATAACAAT	4926
		TCACGACCTAACAATTG	4927
		CAATTGTT <u>A</u> GGTCGTGA	4928

Male-sterile	GGGGCGGGGAAGATTGAGATAAAGCGGATCGAGAACGCCACCA	4929
1210-310-110 123	ACAGGCAGGTGACCTAGTCCAAGCGCCGGTCGGGGATCATGAAG	
riticum aestivum	AAGGCGCGGGAGCTCACCGTGCTCTGCGACGCC	
yr21Term	GGCGTCGCAGAGCACGGTGAGCTCCCGCGCCTTCTTCATGATCC	4930
AC-TAG	CCGACCGGCGCTTGGACTAGGTCACCTGCCTGTTGGTGGCGTTC	
	TCGATCCGCTTTATCTCAATCTTCCCCCGCCCC	
	GTGACCTAGTCCAAGCG	4931
	CGCTTGGA <u>C</u> TAGGTCAC	4932
Male-sterile	CGGGGAAGATTGAGATAAAGCGGATCGAGAACGCCACCAACAG	4933
NP3	GCAGGTGACCTACTCCTAGCGCCGGTCGGGGATCATGAAGAAGG	
Triticum aestivum	CGCGGGAGCTCACCGTGCTCTGCGACGCCCAGG	
ys23Term	CCTGGGCGTCGCAGAGCACGGTGAGCTCCCGCGCCTTCTTCATG	4934
AAG-TAG	ATCCCGACCGGCGCTAGGAGTAGGTCACCTGCCTGTTGGTGGC	
	GTTCTCGATCCGCTTTATCTCAATCTTCCCCCG	
	CCTACTCC <u>T</u> AGCGCCGG	4935
	CCGGCGCT <u>A</u> GGAGTAGG	4936
Male-sterile	TTGAGATAAAGCGGATCGAGAACGCCACCAACAGGCAGGTGACCT	4937
AP3	ACTCCAAGCGCCGGTAGGGGATCATGAAGAAGGCGCGGGAGCTC	
Triticum aestivum	ACCGTGCTCTGCGACGCCCAGGTCGCCATCAT	
Ser26Term	ATGATGGCGACCTGGGCGTCGCAGAGCACGGTGAGCTCCCGCGC	4938
TCG-TAG	CTTCTTCATGATCCCCTACCGGCGCTTGGAGTAGGTCACCTGCCT	
100 11.0	GTTGGTGGCGTTCTCGATCCGCTTTATCTCAA	
	GCGCCGGT <u>A</u> GGGGATCA	4939
	TGATCCCC <u>T</u> ACCGGCGC	4940
Male-sterile	CGGATCGAGAACGCCACCAACAGGCAGGTGACCTACTCCAAGCG	4941
AP3	CCGGTCGGGGATCATG <u>T</u> AGAAGGCGCGGGAGCTCACCGTGCTCT	
Triticum aestivum	GCGACGCCAGGTCGCCATCATCATGTTCTCCT	_
Lys30Term	AGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACGGTG	4942
AAG-TAG	AGCTCCCGCGCCTTCTACATGATCCCCGACCGGCGCTTGGAGTA	
, 0.0 , , .0	GGTCACCTGCCTGTTGGTGGCGTTCTCGATCCG	
	GGATCATG <u>T</u> AGAAGGCG	4943
	CGCCTTCT <u>A</u> CATGATCC	4944
Male-sterile	GGGGCGCGGCAAGATCGAGATCAAGCGGATCGAGAACGCCACCA	4945
Silky1	ACCGCCAGGTGACCTAGTCCAAGCGCCGGACGGGGATCATGAAG	
Zea mays	AAGGCACGCGAGCTCACCGTGCTCTGCGACGCC	
Tyr21Term	GGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATGATCC	4946
TAC-TAG	CCGTCCGGCGCTTGGACTAGGTCACCTGGCGGTTGGTGGCGTTC	
	TCGATCCGCTTGATCTCGATCTTGCCGCGCCCC	
	GTGACCTA <u>G</u> TCCAAGCG	4947
	CGCTTGGACTAGGTCAC	4948

Male-sterile Silky1 CCAGGTGACCTACTCCTAGCGCGGACGGGATCATGAAGAAGG Zea mays Lys23Term AAG-TAG AAG-TAG Male-sterile CGCGGCAAGATCGAGATCAAGCGGATCGAGAACGCCACCAACCG CCAGGTGACCTACTCCTAGCGCCCAGG CCAGGCGCCCAGG CCTGGGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATG ATCCCCGTCCGGCGCTAGGAGTCACCTGGCGGTTGGTGGC CCTACTCCTAGCGCCGG CCTACTCCTAGCGCCGG Male-sterile CGGATCGAGAACGCCACCAACCGCCAGGTGACCTACTCCAAGCG	4949 4950 4951 4952 4953
Zea mays Lys23Term AAG-TAG CCTGGGCGTCGCAGGCTCACCGTGCTCTGCGACGCCCAGG CCTGGGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATG ATCCCCGTCCGGCGCTAGGAGTAGGTCACCTGGCGGTTGGTGGC GTTCTCGATCCGCTTGATCTCGATCTTGCCGCG CCTACTCCTAGCGCCGG CCGGCGCTAGGAGTAGG	4951 4952
Lys23Term CCTGGGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATG AAG-TAG ATCCCCGTCCGGCGCTAGGAGTAGGTCACCTGGCGGTTGGTGGC GTTCTCGATCCGCTTGATCTCGATCTTGCCGCG CCTACTCCTAGCGCCGG CCGGCGCTAGGAGTAGG	4951 4952
AAG-TAG ATCCCCGTCCGGCGCTAGGAGTAGGTCACCTGGCGGTTGGTGGC GTTCTCGATCCGCTTGATCTCGATCTTGCCGCG CCTACTCCTAGCGCCGG CCGGCGCTAGGAGTAGG	4951 4952
GTTCTCGATCCGCTTGATCTCGATCTTGCCGCG CCTACTCCTAGCGCCGG CCGGCGCTAGGAGTAGG	4952
CCTACTCC <u>T</u> AGCGCCGG CCGGCGCT <u>A</u> GGAGTAGG	4952
Male-sterile ICGGATCGAGAACGCCACCAACCGCCAGGTGACCTACTCCAAGCG	4953
Silky1 CCGGACGGGGATCATG <u>T</u> AGAAGGCACGCGAGCTCACCGTGCTCT	
Zea mays GCGACGCCCAGGTCGCCATCATCTCCCT	
Lys30Term AGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACGGTG	4954
10 AAG-TAG AGCTCGCGTGCCTTCTACATGATCCCCGTCCGGCGCTTGGAGTAG	
GTCACCTGGCGGTTGGTGGCGTTCTCGATCCG	
GGATCATG <u>T</u> AGAAGGCA	4955
TGCCTTCT <u>A</u> CATGATCC	4956
Male-sterile ATCGAGAACGCCACCAACCGCCAGGTGACCTACTCCAAGCGCCG	4957
Silky1 GACGGGGATCATGAAG <u>T</u> AGGCACGCGAGCTCACCGTGCTCTGCG	
Zea mays ACGCCCAGGTCGCCATCATCATGTTCTCCTCCA	
Lys31Term TGGAGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACG	4958
15 AAG-TAG GTGAGCTCGCGTGCCTACTTCATGATCCCCGTCCGGCGCTTGGA	
GTAGGTCACCTGGCGGTTGGTGGCGTTCTCGAT	
TCATGAAG <u>T</u> AGGCACGC	4959
GCGTGCCT <u>A</u> CTTCATGA	4960
Male-sterile GCTAGCTGCATTGTCCGGCGAGAGAGATAGCTGCTGCAGGGGGC	4961
AP3 GGCCATGGGGAGGGGC <u>T</u> AGATCGAGATCAAGCGGATCGAGAACG	
Oryza sativa CGACCAACAGGCAGGTGACCTACTCGAAGCGCC	
Lys5Term GGCGCTTCGAGTAGGTCACCTGCCTGTTGGTCGCGTTCTCGATCC	4962
20 AAG-TAG GCTTGATCTCGATCTAGCCCCCATGGCCGCCCCCTGCAGC	
AGCTATCTCTCGCCGGACAATGCAGCTAGC	1000
GGAGGGC <u>T</u> AGATCGAG	4963
CTCGATCT <u>A</u> GCCCCTCC	4964
Male-sterile TGCATTGTCCGGCGAGAGAGATAGCTGCTGCAGGGGGCGCCAT	4965
AP3 GGGGAGGGCAAGATC <u>T</u> AGATCAAGCGGATCGAGAACGCGACCA	
Oryza sativa ACAGGCAGGTGACCTACTCGAAGCGCCGCACGG	
Glu7Term CCGTGCGGGCTTCGAGTAGGTCACCTGCCTGTTGGTCGCGTTCT	4966
25 GAG-TAG CGATCCGCTTGATCTAGATCTTGCCCCTCCCCATGGCCGCCCCCT	
GCAGCAGCTATCTCTCGCCGGACAATGCA	400=
GCAAGATC <u>T</u> AGATCAAG	4967
CTTGATCT <u>A</u> GATCTTGC	4968

Male-sterile AP3	GTCCGGCGAGAGATAGCTGCTGCAGGGGGGCGGCCATGGGGA GGGGCAAGATCGAGATC <u>T</u> AGCGGATCGAGAACGCGACCAACAGG CAGGTGACCTACTCGAAGCGCCGCACGGGGATCA	4969
Oryza sativa Lys9Term AAG-TAG	TGATCCCGTGCGCGCGCTTCGAGTAGGTCACCTGCCTGTTGGTC GCGTTCTCGATCCGCTAGATCTCGATCTTGCCCCCTCCCCATGGCC GCCCCTGCAGCAGCTATCTCTCTCGCCGGAC	4970
	TCGAGATC <u>T</u> AGCGGATC	4971
	GATCCGCT A GATCTCGA	4972
Male-sterile AP3	GAGAGATAGCTGCTGCAGGGGGGGCCATGGGGAGGGGCAAGA TCGAGATCAAGCGGATCTAGAACGCGACCAACAGGCAGGTGACCT ACTCGAAGCGCCGCACGGGGATCATGAAGAAGG	4973
Oryza sativa Glu12Term GAG-TAG	CCTTCTTCATGATCCCCGTGCGGCGCTTCGAGTAGGTCACCTGCC TGTTGGTCGCGTTCTAGATCCGCTTGATCTCGATCTTGCCCCTCC CCATGGCCGCCCCCTGCAGCAGCTATCTCTC	4974
	AGCGGATC <u>T</u> AGAACGCG	4975
	CGCGTTCTAGATCCGCT	4976

Table 15
Oligonucleotides to produce male-sterile plants

15	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
20	Male-sterile AG Arabidopsis thaliana Tyr35Term TAC-TAG	TCTGTACTAATCAAATTTTGCCCTAAACGTTTTTGGCTTTGGAGCA GCAATCACGGCGTAGCAATCGGAGCTAGGAGGAGATTCCTCTCC CTTGAGGAAATCTGGGAGAGGAAAGATCGAA TTCGATCTTTCCTCTCCCAGATTTCCTCAAGGGAGAGGAATCTCCT CCTAGCTCCGATTGCTACGCCGTGATTGCTGCTCCAAAGCCAAAA ACGTTTAGGGCAAAATTTGATTAGTACAGA	4977
		ACGGCGTAGCAATCGGA	4979 4980
	Male-sterile AG	TCCGATTGCTACGCCGT CTGTACTAATCAAATTTTGCCCTAAACGTTTTTGGCTTTGGAGCAG CAATCACGGCGTACTAATCGGAGGAGGAGGATTCCTCCCT TCAACGAAATCTCCCACGAAAGATCGAAA	4981
25	Arabidopsis thaliana Gln36Term CAA-TAA	TGAGGAAATCTGGGAGAGGAAAGATCGAAA TTTCGATCTTTCCTCCCAGATTTCCTCAAGGGAGAGGAATCTCC TCCTAGCTCCGATTAGTACGCCGTGATTGCTGCTCCAAAGCCAAA AACGTTTAGGGCAAAATTTGATTAGTACAG	4982
		CGGCGTACTAATCGGAG	4983
		CTCCGATT A GTACGCCG	4984

Phenotype, Gene, Plant & Targeted Afteration	Altering Oligos	SEQ ID NO:
Male-sterile AG Arabidopsis thaliana	ACTAATCAAATTTTGCCCTAAACGTTTTTGGCTTTGGAGCAGCAAT CACGGCGTACCAAT <u>A</u> GGAGCTAGGAGGAGATTCCTCTCCCTTGA GGAAATCTGGGAGAGGGAAAGATCGAAATCAA	4985
Ser37Term TCG-TAG	TTGATTTCGATCTTTCCTCTCCCAGATTTCCTCAAGGGAGGAAT CTCCTCCTAGCTCCTATTGGTACGCCGTGATTGCTGCTCCAAAGC CAAAAACGTTTAGGGCAAAATTTGATTAGT	4986
	GTACCAAT <u>A</u> GGAGCTAG	4987
	CTAGCTCC <u>T</u> ATTGGTAC	4988
Male-sterile AG Arabidopsis thaliana	TAATCAAATTTTGCCCTAAACGTTTTTGGCTTTGGAGCAGCAATCA CGGCGTACCAATCGTAGCTAGGAGGAGATTCCTCTCCCTTGAGGA AATCTGGGAGAGGAAAGATCGAAATCAAAC	4989
Glu38Term GAG-TAG	GTTTGATTTCGATCTTTCCTCTCCCAGATTTCCTCAAGGGAGAGGA ATCTCCTCCTAGCTACGATTGGTACGCCGTGATTGCTGCTCCAAA GCCAAAAACGTTTAGGGCAAAATTTGATTA	4990
	ACCAATCG <u>T</u> AGCTAGGA	4991
	TCCTAGCT <u>A</u> CGATTGGT	4992
Male-sterile AG Brassica napus	CTCTCCCACTTCTTTTCGGTGGTTTATTCATTTGGTGACGATATCA CAGAAGCAATGGATTAAGGTGGGAGTAGTCACGATGCAGAGAGTA GCAAGAAGATAGGTAGAGGGAAGATAGAGA	4993
Glu3Term GAA-TAA	TCTCTATCTTCCCTCTACCTATCTTCTTGCTACTCTCTGCATCGTG ACTACTCCCACCTTAATCCATTGCTTCTGTGATATCGTCACCAAAT GAATAAACCACCGAAAAGAAGTGGGAGAG	4994
	CAATGGAT <u>T</u> AAGGTGGG	4995
	CCCACCTT <u>A</u> ATCCATTG	4996
Male-sterile AG Brassica napus	TATTCATTTGGTGACGATATCACAGAAGCAATGGATGAAGGTGGG AGTAGTCACGATGCA <u>T</u> AGAGTAGCAAGAAGATAGGTAGAGGGAAG ATAGAGATAAAGAGGATAGAGAACACAA	4997
Glu11Term GAG-TAG	TTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTCCCTCTACCTATC TTCTTGCTACTCTATCCATCGTGACTACTCCCACCTTCATCCATTG CTTCTGTGATATCGTCACCAAATGAATA	4998
	ACGATGCA <u>T</u> AGAGTAGC	4999
	GCTACTCT <u>A</u> TGCATCGT	5000
Male-sterile AG Brassica napus	GGTGACGATATCACAGAAGCAATGGATGAAGGTGGGAGTAGTCA CGATGCAGAGAGTAGCTAGAAGATAGGTAGAGGAAGATAGAGAT AAAGAGGATAGAGAACACAAATCGTCAAG	5001
Lys14Term AAG-TAG	CTTGACGATTTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTCCCT CTACCTATCTTCTAGCTACTCTCTGCATCGTGACTACTCCCACCTT CATCCATTGCTTCTGTGATATCGTCACC	5002
	AGAGTAGC <u>T</u> AGAAGATA	5003
	TATCTTCTAGCTACTCT	5004

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile AG Brassica napus	GACGATATCACAGAAGCAATGGATGAAGGTGGGAGTAGTCACGAT GCAGAGAGTAGCAAGTAGATAGGTAGAGGGAAGATAGAGATAAAG AGGATAGAGAACAAACAAATCGTCAAGTAA	5005
Lys15Term AAG-TAG	TTACTTGACGATTTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTC CCTCTACCTATCTACTTGCTACTCTCTGCATCGTGACTACTCCCAC CTTCATCCATTGCTTCTGTGATATCGTC	5006
	GTAGCAAG <u>T</u> AGATAGGT	5007
	ACCTATCT <u>A</u> CTTGCTAC	5008
Male-sterile AG Lycopersicon	CAACCAAAAACTTAAAAATCTTCTCTTTCCTTTCCTTACAAGGTGA AGTAATGGACTTC <u>T</u> AAAGTGATCTAACCAGAGAGATCTCACCACAA AGGAAACTAGGAAGGGGGAAAATTGAGA	5009
esculentum Glu4Term CAA-TAA	TCTCAATTTCCCCCTTCCTAGTTTCCTTTGTGGTGAGATCTCTCT GGTTAGATCACTTTAGAAGTCCATTACTTCACCTTGTAAGGAAAGG AAAGAGAAGATTTTTAAGTTTTTTGGTTG	5010
OAA-1AA	TGGACTTC <u>T</u> AAAGTGAT	5011
	ATCACTTT <u>A</u> GAAGTCCA	5012
Male-sterile AG	AAAATCTTCTCTTTCCTTTCCTTACAAGGTGAAGTAATGGACTTCC AAAGTGATCTAACCTGAGAGATCTCACCACAAAGGAAACTAGGAA GGGGGAAAATTGAGATCAAAAGGATCGAAA	5013
Lycopersicon esculentum Arg9Term AGA-TGA	TTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTAGTTTCCTTTGT GGTGAGATCTCTCAGGGTAGATCACTTTGGAAGTCCATTACTTCAC CTTGTAAGGAAAGGA	5014
AGA-TGA	ATCTAACCTGAGAGATC	5015
	GATCTCTC <u>A</u> GGTTAGAT	5016
Male-sterile AG	ATCTTCTCTTTCCTTTCCTTACAAGGTGAAGTAATGGACTTCCAAA GTGATCTAACCAGATAGATCTCACCACAAAGGAAACTAGGAAGGG GGAAAATTGAGATCAAAAGGATCGAAAACA	5017
Lycopersicon esculentum Glu10Term GAG-TAG	TGTTTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTAGTTTCCTT TGTGGTGAGATCTATCTGGTTAGATCACTTTGGAAGTCCATTACTT CACCTTGTAAGGAAAGGA	5018
GAG-TAG	TAACCAGATAGATCTCA	5019
	TGAGATCT <u>A</u> TCTGGTTA	5020
Male-sterile AG	CTTTCCTTTCCAAGGTGAAGTAATGGACTTCCAAAGTGATCT AACCAGAGAGATCTGACCACAAAGGAAACTAGGAAGGGGGAAAAT TGAGATCAAAAGGATCGAAAACACGACGAA	5021
Lycopersicon esculentum Ser12Term TCA-TGA	TTCGTCGTGTTTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTA GTTTCCTTTGTGGTCAGATCTCTCTGGTTAGATCACTTTGGAAGTC CATTACTTCACCTTGTAAGGAAAGGA	5022
10/4-10/1	AGAGATCTGACCACAAA	5023

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TTTGTGGT <u>C</u> AGATCTCT	5024
Male-sterile NAG1 Nicotiana tabacum	GTACTCTCTATTTTCATCTTCCAACCCTTTCTTTCCTTACCAGGTGA AAGTATGGACTTCTAAAGTGATCTAACAAGAGAGATCTCTCCACAA AGGAAACTGGGAAGAGAGAAGATTGAGA	5025
Gln4Term CAA-TAA	TCTCAATCTTTCCTCTTCCCAGTTTCCTTTGTGGAGAGATCTCTCT TGTTAGATCACTTTAGAAGTCCATACTTTCACCTGGTAAGGAAAGA AAGGGTTGGAAGATGAAAATAGAGAGTAC	5026
	TGGACTTC <u>T</u> AAAGTGAT	5027
	ATCACTIT A GAAGTCCA	5028
Male-sterile NAG1 Nicotiana tabacum	ATCTTCCAACCCTTTCTTTCCTTACCAGGTGAAAGTATGGACTTCC AAAGTGATCTAACATGAGAGATCTCTCCACAAAGGAAACTGGGAA GAGGAAAGATTGAGATCAAACGGATCGAAA	5029
Arg9Term AGA-TGA	TTTCGATCCGTTTGATCTCAATCTTTCCTCTTCCCAGTTTCCTTTGT GGAGAGATCTCTCACTTAGATCACTTTGGAAGTCCATACTTTCAC CTGGTAAGGAAAGAAAGGGTTGGAAGAT	5030
	ATCTAACA <u>T</u> GAGAGATC	5031
	GATCTCTC <u>A</u> TGTTAGAT	5032
Male-sterile NAG1 Nicotiana tabacum	TTCCAACCCTTTCTTTCCTTACCAGGTGAAAGTATGGACTTCCAAA GTGATCTAACAAGA <u>T</u> AGATCTCTCCACAAAGGAAACTGGGAAGAG GAAAGATTGAGATCAAACGGATCGAAAACA	5033
Glu10Term GAG-TAG	TGTTTTCGATCCGTTTGATCTCAATCTTTCCTCTTCCCAGTTTCCTT TGTGGAGAGATCTACTTGTTAGATCACTTTGGAAGTCCATACTTT CACCTGGTAAGGAAAGAAAGGGTTGGAA	5034
•	TAACAAGA <u>T</u> AGATCTCT	5035
	AGAGATCT <u>A</u> TCTTGTTA	5036
Male-sterile NAG1 Nicotiana tabacum	CTITCCTTACCAGGTGAAAGTATGGACTTCCAAAGTGATCTAACAA GAGAGATCTCTCCATAAAGGAAACTGGGAAGAGGAAAGATTGAGA TCAAACGGATCGAAAACACAACGAATCGTC	5037
Gln14Term CAA-TAA	GACGATTCGTTGTGTTTTCGATCCGTTTGATCTCAATCTTTCCTCT TCCCAGTTTCCTTTATGGAGAGATCTCTCTTGTTAGATCACTTTGG AAGTCCATACTTTCACCTGGTAAGGAAAG	5038
	TCTCTCCA <u>T</u> AAAGGAAA	5039
	TTTCCTTT A TGGAGAGA	5040
Male-sterile AG Rosa hybrida	GCCTATGAAAACAAACCCAACACGGTCCTGGACGCTGATGCCCAA AGAAGATTGGGAAGGTGAAAGATCGAGATCAAGCGGATCGAAAAC ACCACCAATCGTCAAGTCACCTTCTGCAAAA	5041
Gly22Term GGA-TGA	TTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTTTCGATCCGCT TGATCTCGATCTTTCACCTTCCCAATCTTCTTTGGGCATCAGCGTC CAGGACCGTGTTGGGTTTGTTTTCATAGGC	5042
	TGGGAAGGTGAAAGATC	5043
	GATCTITC <u>A</u> CCTTCCCA	5044

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile AG Rosa hybrida	TATGAAAACAAACCCAACACGGTCCTGGACGCTGATGCCCAAAGA AGATTGGGAAGGGGA <u>T</u> AGATCGAGATCAAGCGGATCGAAAACAC CACCAATCGTCAAGTCACCTTCTGCAAAAGGC	5045
Lys23Term AAG-TAG	GCCTTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTTTCGATCC GCTTGATCTCGATCTATCCCCTTCCCAATCTTCTTTGGGCATCAGC GTCCAGGACCGTGTTGGGTTTGTTTTCATA	5046
	GAAGGGA <u>T</u> AGATCGAG	5047
	CTCGATCT <u>A</u> TCCCCTTC	5048
Male-sterile AG Rosa hybrida	AACAAACCCAACACGGTCCTGGACGCTGATGCCCAAAGAAGATTG GGAAGGGGAAAGATC <u>T</u> AGATCAAGCGGATCGAAAACACCACCAAT CGTCAAGTCACCTTCTGCAAAAGGCGCAATG	5049
Glu25Term GAG-TAG	CATTGCGCCTTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTTT CGATCCGCTTGATCTAGATCTTTCCCCTTCCCAATCTTCTTTGGGC ATCAGCGTCCAGGACCGTGTTGGGTTTGTT	5050
	GAAAGATCTAGATCAAG	5051
	CTTGATCTAGATCTTTC	5052
Male-sterile AG	CCCAACACGGTCCTGGACGCTGATGCCCAAAGAAGATTGGGAAG GGGAAAGATCGAGATCTAGCGGATCGAAAACACCACCAATCGTCA AGTCACCTTCTGCAAAAGGCGCAATGGTTTGC	5053
Rosa hybrida Lys27 AAG-TAG	GCAAACCATTGCGCCTTTTGCAGAAGGTGACTTGACGATTGGTGG TGTTTTCGATCCGCTAGATCTCGATCTTTCCCCTTCCCAATCTTCT TTGGGCATCAGCGTCCAGGACCGTGTTGGG	5054
	TCGAGATCTAGCGGATC	505
	GATCCGCTAGATCTCGA	505
Male-sterile far	CAATTGCCTGTTTTATTTTTTTTTTTTTTTGACTAAGTAGAAATGGC GTCTCTAAGCGATTAATCGACCGAGGTATCGCCCGAGAGGAAAAT CGGGAGAGGAAAGATCGAGATCAAACGGA	505
Antirrhinum majus Gln7Term CAA-TAA	TCCGTTTGATCTCGATCTTTCCTCTCCCGATTTTCCTCTCGGGCGA TACCTCGGTCGATTAATCGCTTAGAGACGCCATTTCTACTTAGTCA AAAAGAAAAAAAAAA	505
	TAAGCGATTAATCGACC	505
	GGTCGATT <u>A</u> ATCGCTTA	506
Male-sterile far Antirrhinum majus	GTTTTTATTTTTTCTTTTTGACTAAGTAGAAATGGCGTCTCTAAG CGATCAATCGACCTAGGTATCGCCCGAGAGGAAAATCGGGAGAG GAAAGATCGAGATCAAACGGATCGAAAACA	506
Glu10Term GAG-TAG	TGTTTTCGATCCGTTTGATCTCGATCTTTCCTCTCCCGATTTTCCT CTCGGGCGATACCTAGGGTCGATTGATCGCTTAGAGACGCCATTTC TACTTAGTCAAAAAGAAAAAAAAAA	506
	AATCGACCTAGGTATCG	506
	CGATACCTAGGTCGATT	506

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Male-sterile far Antirrhinum majus	TTTCTTTTTGACTAAGTAGAAATGGCGTCTCTAAGCGATCAATCGA CCGAGGTATCGCCC <u>T</u> AGAGGAAAATCGGGAGAGGAAAGATCGAG ATCAAACGGATCGAAAACAAAAC	5065
Glu14Term GAG-TAG	GTTGATTTGTTTTCGATCCGTTTGATCTCGATCTTTCCTCTC CCGATTTTCCTCTAGGGGCGATACCTCGGTCGATTGATCGCTTAGA GACGCCATTTCTACTTAGTCAAAAAGAAA	5066
•	TATCGCCCTAGAGGAAA	5067
	TTTCCTCT <u>A</u> GGGCGATA	5068
Male-sterile far <i>Antirrhinum majus</i>	TTTGACTAAGTAGAAATGGCGTCTCTAAGCGATCAATCGACCGAG GTATCGCCCGAGAGG <u>T</u> AAATCGGGAGAGGAAAGATCGAGATCAA ACGGATCGAAAACAAAAC	5069
Lys16Term AAA-TAA	TAACCTGTTGATTTGTTTTGTTTTCGATCCGTTTGATCTCGATCTTT CCTCTCCCGATTTACCTCGGGCGATACCTCGGTCGATTGATCG CTTAGAGACGCCATTTCTACTTAGTCAAA	5070
	CCGAGAGG <u>T</u> AAATCGGG	5071
	CCCGATTT <u>A</u> CCTCTCGG	5072
Male-sterile AG Cucumis sativus	TGTCCAAGCATTATCAGTCACCACTCACAAGAATGATTAAGGAAGA AGGAAAGGGTAAGTAGCAAATAAAGGGGATGTTCCAGAATCAAGA AGAGAAGATGTCAGACTCGCCTCAGAGGAA	5073
Leu21Term TTG-TAG	TTCCTCTGAGGCGAGTCTGACATCTTCTTCTTGATTCTGGAACA TCCCCTTTATTTGCTACTTACCCTTTCCTTCCTTAATCATTCTT GTGAGTGGTGACTGATAATGCTTGGACA	5074
	GGGTAAGT A GCAAATAA	5075
	TTATTTGC <u>T</u> ACTTACCC	5076
Male-sterile AG Cucumis sativus	TCCAAGCATTATCAGTCACCACTCACAAGAATGATTAAGGAAGAAG GAAAGGGTAAGTTG <u>T</u> AAATAAAGGGGATGTTCCAGAATCAAGAAG AGAAGATGTCAGACTCGCCTCAGAGGAAGA	5077
GIn22Term CAA-TAA	TCTTCCTCTGAGGCGAGTCTGACATCTTCTTCTTGATTCTGGAA CATCCCCTTTATTTACAACTTACCCTTTCCTTCCTTAATCATTC TTGTGAGTGGTGACTGATAATGCTTGGA	5078
	GTAAGTTG <u>T</u> AAATAAAG	5079
	CTTTATTT <u>A</u> CAACTTAC	5080
Male-sterile AG Cucumis sativus	CATTATCAGTCACCACTCACAAGAATGATTAAGGAAGAAGGAAAG GGTAAGTTGCAAATA <u>T</u> AGGGGATGTTCCAGAATCAAGAAGAAGAAG ATGTCAGACTCGCCTCAGAGGAAGATGGGAA	5081
Lys24Term AAG-TAG	TTCCCATCTTCCTGAGGCGAGTCTGACATCTTCTCTTGATT CTGGAACATCCCCTATATTTGCAACTTACCCTTTCCTTCTTA ATCATTCTTGTGAGTGGTGACTGATAATG	5082
	TGCAAATA <u>T</u> AGGGGATG	5083
	CATCCCCTATATITGCA	5084

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile AG Cucumis sativus	CCACTCACAAGAATGATTAAGGAAGAAGGAAAGGGTAAGTTGCAA ATAAAGGGGATGTTCTAGAATCAAGAAGAAGATGTCAGACTCG CCTCAGAGGAAGATGGGAAAGAGAGATTG	5085
GIn28Term CAG-TAG	CAATCTTTCCTCTTCCCATCTTCCTCTGAGGCGAGTCTGACATCTT CTCTTCTTGATTCTAGAACATCCCCTTTATTTGCAACTTACCCTTTC CTTCTTCCTTAATCATTCTTGTGAGTGG	5086
	GGATGTTCTAGAATCAA	5087
	TTGATTCTAGAACATCC	5088
Male-sterile AG Zea mays	CCACCACCACCACCACCACCACCACCATGCTCAACATGAT GACTGATCTGAGCTGAG	5089
Cys10Term TGC-TGA	CCTGTCGCCGGAGCCCGTCGGCGCCGCCGCCACCTGCTCCTTG ACCTTGGACGACGGCCCTCAGCTCAG	5090
	CTGAGCTGAGGCCGTC .	5091
	GACGGCCC <u>T</u> CAGCTCAG	5092
Male-sterile AG	ACCACCACCACCACCACCACCATGCTCAACATGATGACTGATC TGAGCTGCGGGCCGTAGTCCAAGGTCAAGGAGCAGGTGGCGGC GGCGCCGACGGGCTCCGGCGACAGGCAGGGGCA	5093
Zea mays Ser13Term TCG-TAG	TGCCCTGCCTGTCGCCGGAGCCCGTCGGCGCCGCCACCT GCTCCTTGACCTTGGACTACGGCCCGCAGCTCAGATCAGTCATCA TGTTGAGCATGGTGTGGTG	5094
	CGGGCCGTAGTCCAAGG	5095
	CCTTGGACTACGGCCCG	5096
Male-sterile AG Zea mays	CACCACCACCACCACCATGCTCAACATGATGACTGATCTGAGC TGCGGGCCGTCGTCCTAGGTCAAGGAGCAGGTGGCGGCGCGCCCCGACGGGCTCCGGCGACAGGCAGG	5097
Lys15Term AAG-TAG	TCCCTGCCCTGCCTGTCGCCGGAGCCCGTCGGCGCCGCCGCCGCCGCCGCCGCTGCTCCTTGACCTAGGACGACGGCCCGCAGCTCAGATCAGTCATCATGTTGAGCATGGTGTGGTGGTGGTGGTGGTG	5098
	CGTCGTCCTAGGTCAAG	509
	CTTGACCT <u>A</u> GGACGACG	510
Male-sterile AG Zea mays	CACCACCACACCATGCTCAACATGATGACTGATCTGAGCTGCGGG CCGTCGTCCAAGGTCTAGGAGCAGGTGGCGGCGCGCCGACGG GCTCCGGCGACAGGCAGGGGAGAGGCA	510
Lys17Term AAG-TAG	TGCCTCTCCCTGCCTGCCTGTCGCCGAGCCCGTCGGCGC CGCCGCCACCTGCTCCTAGACCTTGGACGACGCCCGCAGCTCA GATCAGTCATCATGTTGAGCATGGTGTGGTG	
	CCAAGGTCTAGGAGCAG	510
	CTGCTCCTAGACCTTGG	510

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Male-sterile AG Zea mays	TCCTACCTTTCTCCTTCAGACCTCAAAATCTGTGTGATAGGAACA AGAGCATGCACATC <u>T</u> GAGAAGAGGGGGCTACACCATCCACAGTAA CAGGCATCATGTCGACCCTGACTTCGGCGG	5105
Arg4Term CGA-TGA	CCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTGGATGGT GTAGCCTCCTCTTCTCAGATGTGCATGCTCTTGTTCCTATCACACA GATTTTGAGGTCTGAAGGAGAAAAGGTAGGA	5106
	TGCACATCTGAGAAGAG	5107
	CTCTTCTC <u>A</u> GATGTGCA	5108
Male-sterile AG Zea mays	TACCTTTTCTCCTTCAGACCTCAAAATCTGTGTGATAGGAACAAGA GCATGCACATCCGA <u>T</u> AAGAGGAGGCTACACCATCCACAGTAACAG GCATCATGTCGACCCTGACTTCGGCGGGGC	5109
Glu5Term GAA-TAA	GCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTGGAT GGTGTAGCCTCCTCTTATCGGATGTGCATGCTCTTGTTCCTATCA CACAGATTTTGAGGTCTGAAGGAGAAAAGGTA	5110
	ACATCCGA T AAGAGGAG	5111
	CTCCTCTTATCGGATGT	5112
Male-sterile AG Zea mays	CTTTTCTCCTTCAGACCTCAAAATCTGTGTGATAGGAACAAGAGCA TGCACATCCGAGAATAGGAGGCTACACCATCCACAGTAACAGGCA TCATGTCGACCCTGACTTCGGCGGGGCAGC	5113
Glu6Term GAG-TAG	GCTGCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTG GATGGTGTAGCCTCCTATTCTCGGATGTGCATGCTCTTGTTCCTA TCACACAGATTTTGAGGTCTGAAGGAGAAAAG	5114
	TCCGAGAA <u>T</u> AGGAGGCT	5115
	AGCCTCCT <u>A</u> TTCTCGGA	5116
Male-sterile AG Zea mays	TTCTCCTTCAGACCTCAAAATCTGTGTGATAGGAACAAGAGCATGC ACATCCGAGAAGAGTAGGCTACACCATCCACAGTAACAGGCATCA TGTCGACCCTGACTTCGGCGGGGCAGCAGA	5117
Glu7Term GAG-TAG	TCTGCTGCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACT GTGGATGGTGTAGCCT <u>A</u> CTCTTCTCGGATGTGCATGCTCTTGTTC CTATCACACAGATTTTGAGGTCTGAAGGAGAA	5118
	GAGAAGAG <u>T</u> AGGCTACA	5119
	TGTAGCCT <u>A</u> CTCTTCTC	5120
Male-sterile AG O <i>ryza sativa</i>	GCTGGGTCAGGATCGTCGGCGGCGGGGGGGGGGGGGGGG	5121
Lys5Term AAG-TAG	GGCGCTTGCAGAAGGTCACCTGCCGGTTCGTCGTGTTCTCGATC CGCTTTATCTCGATCTACCCCCTCCCCATCTTCTCGCTGCTCCCC GCCGCCACCGCCGCCGACGATCCTGACCCAGC	5122
	GGAGGGG <u>T</u> AGATCGAG	5123
	CTCGATCTACCCCCTCC	5124

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Male-sterile AG <i>Oryza sativa</i>	TCAGGATCGTCGGCGCGGGGGGGGGGGAGAAGA TGGGGAGGGGGAAGATC <u>T</u> AGATAAAGCGGATCGAGAACACGACG AACCGGCAGGTGACCTTCTGCAAGCGCCGCAATG	5125
Glu7Term GAG-TAG	CATTGCGGCGCTTGCAGAAGGTCACCTGCCGGTTCGTCGTTC TCGATCCGCTTTATCTAGATCTTCCCCCTCCCCATCTTCTCGCTG CTCCCCGCCGCCACCGCCGCCGACGATCCTGA	5126
	GGAAGATC <u>T</u> AGATAAAG	5127
	CTTTATCTAGATCTTCC	5128
Male-sterile AG	TCGTCGGCGCGGTGGCGGCGGGGAGCAGCGAGAAGATGGGG AGGGGGAAGATCGAGATATAGCGGATCGAGAACACGACGAACCG GCAGGTGACCTTCTGCAAGCGCCGCAATGGCCTCC	5129
Oryza sativa Lys9Term AAG-TAG	GGAGGCCATTGCGGCGCTTGCAGAAGGTCACCTGCCGGTTCGTC GTGTTCTCGATCCGCTATATCTCGATCTTCCCCCCTCCCCATCTTCT CGCTGCTCCCCGCCGCCGCCGCCGACGA	5130
	TCGAGATATAGCGGATC	5131
	GATCCGCT <u>A</u> TATCTCGA	5132
Male-sterile AG Oryza sativa	GCGGTGGCGGGGGGGGGGGGGGGGGGGGGGGGGGGGGG	5133
Glu12Term GAG-TAG	CCTTCTTCAGGAGGCCATTGCGGCGCTTGCAGAAGGTCACCTGC CGGTTCGTCGTGTTCTAGATCCGCTTTATCTCGATCTTCCCCCTC CCCATCTTCTCGCTGCTCCCCGCCGCCACCGC	5134
	AGCGGATCTAGAACACG	5135
	CGTGTTCT <u>A</u> GATCCGCT	5136

Table 16
Oligonucleotides to produce male-sterile plants

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile	GGGAAGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAAT AGACAAGTTACATAGTCAAAGAGAAGAAATGGTATCATCAAAAAAG	5137
Cucumis sativus	CCAAAGAAATTACTGTTCTTTGCGATGCT	
Tyr21Term TAT-TAG	AGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTTGATGATACCAT TTCTTCTCTTTGACTATGTAACTTGTCTATTGCTTGAGTTCTCTATTC TTTTTATTTCTATTTTCCCTCTTCCC	5138

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GTTACATAGTCAAAGAG	5139
	CTCTTTGA <u>C</u> TATGTAAC	5140
Male-sterile PI Cucumis sativus	GAAGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAG ACAAGTTACATATT <u>G</u> AAAGAGAAGAAATGGTATCATCAAAAAAGCC AAAGAAATTACTGTTCTTTGCGATGCTCA	5141
Ser22Term TCA-TGA	TGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTTGATGATACC ATTTCTTCTCTTTCAATATGTAACTTGTCTATTGCTTGAGTTCTCTAT TCTTTTTATTTCTATTTTCCCTCTTC	5142
	TACATATT G AAAGAGAA	5143
	TTCTCTTT <u>C</u> AATATGTA	5144
Male-sterile Pl Cucumis sativus	AGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAGAC AAGTTACATATTCA <u>T</u> AGAGAAGAAATGGTATCATCAAAAAAAGCCAAA GAAATTACTGTTCTTTGCGATGCTCAAG	5145
Lys23Term AAG-TAG	CTTGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTGATGATA CCATTTCTTCTCTATGAATATGTAACTTGTCTATTGCTTGAGTTCTC TATTCTTTTTATTTCTATTTTCCCTCT	5146
	CATATTCA <u>T</u> AGAGAAGA	5147
	TCTTCTCT <u>A</u> TGAATATG	5148
Male-sterile Pl Cucumis sativus	GGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAGACAAG TTACATATTCAAAG <u>T</u> GAAGAAATGGTATCATCAAAAAAAGCCAAAGAA ATTACTGTTCTTTGCGATGCTCAAGTTT	5149
Arg24Term AGA-TGA	AAACTTGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTTGATG ATACCATTTCTTCACTTTGAATATGTAACTTGTCTATTGCTTGAGTT CTCTATTCTTTTTATTTCTATTTTCCC	5150
	ATTCAAAG <u>T</u> GAAGAAAT	5151
	ATTTCTTC <u>A</u> CTTTGAAT	5152
Male-sterile Pl Malus domestica	GGGACGTGGGAAGGTTGAGATCAAGAGGATTGAGAACTCAAGTAA CAGGCAGGTGACCTAGTCCAAGAGGAGGAATGGGATTATCAAGAA GGCAAAGGAGATCACTGTTCTATGTGATGCT	5153
Tyr21Term TAC-TAG	AGCATCACATAGAACAGTGATCTCCTTTGCCTTCTTGATAATCCCA TTCCTCCTCTTGGACTAGGTCACCTGCCTGTTACTTGAGTTCTCAA TCCTCTTGATCTCAACCTTCCCACGTCCC	5154
	GTGACCTA <u>G</u> TCCAAGAG	5155
•	CTCTTGGA <u>C</u> TAGGTCAC	5156

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Male-sterile Pl Malus domestica	CGTGGGAAGGTTGAGATCAAGAGGATTGAGAACTCAAGTAACAGG CAGGTGACCTACTCCTAGAGGAGGAATGGGATTATCAAGAAGGCA AAGGAGATCACTGTTCTATGTGATGCTAAAG	5157
Lys23Term AAG-TAG	CTTTAGCATCACATAGAACAGTGATCTCCTTTGCCTTCTTGATAATC CCATTCCTCCTCTAGGAGTAGGTCACCTGCCTGTTACTTGAGTTCT CAATCCTCTTGATCTCAACCTTCCCACG	5158
	CCTACTCC <u>T</u> AGAGGAGG	5159
	CCTCCTCT <u>A</u> GGAGTAGG	5160
Male-sterile Pl Malus domestica	AGGATTGAGAACTCAAGTAACAGGCAGGTGACCTACTCCAAGAGG AGGAATGGGATTATCTAGAAGGCAAAGGAGATCACTGTTCTATGTG ATGCTAAAGTATCTCTTATCATTTATTCTA	5161
Lys30Term AAG-TAG	TAGAATAAATGATAAGAGATACTTTAGCATCACATAGAACAGTGATC TCCTTTGCCTTCTAGATAATCCCATTCCTCCTCTTGGAGTAGGTCA CCTGCCTGTTACTTGAGTTCTCAATCCT	5162
	GGATTATC <u>T</u> AGAAGGCA	5163
	TGCCTTCT <u>A</u> GATAATCC	5164
Male-sterile PI Malus domestica	ATTGAGAACTCAAGTAACAGGCAGGTGACCTACTCCAAGAGGAGG AATGGGATTATCAAG <u>T</u> AGGCAAAGGAGATCACTGTTCTATGTGATG CTAAAGTATCTCTTATCATTTATTCTAGCT	5165
Lys31Term AAG-TAG	AGCTAGAATAAATGATAAGAGATACTTTAGCATCACATAGAACAGT GATCTCCTTTGCCTACTTGATAATCCCATTCCTCCTCTTGGAGTAG GTCACCTGCCTGTTACTTGAGTTCTCAAT	5166
	TTATCAAGTAGGCAAAG	5167
	CTTTGCCTACTTGATAA	5168
Male-sterile globosa	CATTITTACAATAGTTATCTGCAAACAAAAACAAGAGAGAAAAAACAA AAACAAAAAAATG <u>T</u> GAAGAGGAAAAATTGAGATCAAAAGAATTGAG AACTCAAGCAACAGGCAGGTTACTTACT	5169
Antirrhinum majus Gly2Term GGA-TGA	AGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTTGATCTCA ATTTTTCCTCTTCACATTTTTTTGTTTTTGTTTTTCTCTCTTGTTTTTG TTTGCAGATAACTATTGTAAAAATG	5170
	AAAAATG <u>T</u> GAAGAGGA	5171
	TCCTCTTCACATTTTTT	5172
Male-sterile globosa	TTTTACAATAGTTATCTGCAAACAAAAACAAGAGAGAAAAAAAA	5173
Antirrhinum majus Arg3Term AGA-TGA	TTGAGCACAGGCAGGTACTTACTOAN TTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTTGATC TCAATTTTTCCTCATCCCATTTTTTTGTTTTTGTTTTCTCTCTTGTT TTTGTTTGCAGATAACTATTGTAAAA	5174

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
=======================================	AAATGGGA <u>T</u> GAGGAAAA	5175
	TTTTCCTCATCCCATTT	5176
Male-sterile globosa <i>Antirrhinum majus</i>	TACAATAGTTATCTGCAAACAAAACAAGAGAGAAAAAAAA	5177
Gly4Term GGA-TGA	TCTTTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTTG ATCTCAATTTTTCATCTTCCCATTTTTTTGTTTTTGTTTTTCTCTCTT GTTTTTGTTTGCAGATAACTATTGTA	5178
	TGGGAAGA <u>T</u> GAAAAATT	5179
	AATTTTC <u>A</u> TCTTCCCA	5180
Male-sterile globosa <i>Antirrhinum majus</i>	AATAGTTATCTGCAAACAAAACAAGAGAGAAAAACAAAAACAAAAA AATGGGAAGAGGA <u>T</u> AAATTGAGATCAAAAGAATTGAGAACTCAAGC AACAGGCAGGTTACTTACTCAAAGAGAA	5181
Lys5Term AAA-TAA	TTCTCTTTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTT TTGATCTCAATTTATCCTCTTCCCATTTTTTTGTTTTTGTTTTTCTCT CTTGTTTTTGTTTGCAGATAACTATT	5182
	GAAGAGGA <u>T</u> AAATTGAG	5183
	CTCAATTT <u>A</u> TCCTCTTC	5184
Male-sterile Pl Zea mays	GCTGAGCTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGC AGTATGGGGCGCGGCTAGATCAAGATCAAGAGGATCGAGAACTCT ACCAACCGGCAGGTGACCTTCTCCAAGCGCC	5185
Lys5Term AAG-TAG	GGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTCTCGATCC TCTTGATCTTGATCTAGCCGCGCCCCCATACTGCGTTCTCCACTCC CAAACAGATCCAAGGGCAGCAAGAGCTCAGC	5186
	GGCGCGCTAGATCAAG	5187
	CTTGATCT <u>A</u> GCCGCGCC	5188
Male-sterile PI Zea mays	CTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGCAGTATG GGGCGCGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCCG	5189
Lys/Term AAG-TAG	CGGCCCGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTGCGTTCTC CACTCCCAAACAGATCCAAGGGCAGCAAGAG	5190
	GCAAGATC <u>T</u> AGATCAAG	5191
,	CTTGATCT <u>A</u> GATCTTGC	5192

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile	CTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGCAGTATG GGGCGCGGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCCG	5193
Zea mays _ys9Term AAG-TAG	CGGCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTGCGTTCTC CACTCCCAAACAGATCCAAGGGCAGCAAGAG	5194
	GCAAGATC <u>T</u> AGATCAAG	5195
	CTTGATCT A GATCTTGC	5196
Male-sterile Pl Zea mays	GATCTGTTTGGGAGTGGAGAACGCAGTATGGGGCGCGCAAGAT CAAGATCAAGAGGATC <u>T</u> AGAACTCTACCAACCGGCAGGTGACCTT CTCCAAGCGCCGGGCCGG	5197
Glu12Term GAG-TAG	CCTTCTTGACCAGTCCGGCCCGGCGCTTGGAGAAGGTCACCTGC CGGTTGGTAGAGTTCTAGATCCTCTTGATCTTGATCTTGCCGCGC CCCATACTGCGTTCTCCACTCCCAAACAGATC	5198
	AGAGGATC <u>T</u> AGAACTCT	5199
	AGAGTTCT <u>A</u> GATCCTCT	5200
Male-sterile Pl	GCTGAGCTCTTGCTGCCCTTGAATCTGTTAGGGAGTGGAGAACGG AGTATGGGGCGCGGCTAGATCGAGATCAAGAGGATCGAGAACTCT ACCAACCGGCAGGTGACCTTCTCCAAGCGCC	5201
Zea mays Lys5Term AAG-TAG	GGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTCTCGATCC TCTTGATCTCGATCTAGCCGCGCCCCATACTCCGTTCTCCACTCC CTAACAGATTCAAGGGCAGCAAGAGCTCAGC	5202
	GGCGCGCTAGATCGAG	5203
	CTCGATCTAGCCGCGCC	5204
Male-sterile PI	CTCTTGCTGCCCTTGAATCTGTTAGGGAGTGGAGAACGGAGTATG GGGCGCGGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCCG	5205
Zea mays Glu7Term GAG-TAG	CGGCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTCCGTTCTC CACTCCCTAACAGATTCAAGGGCAGCAAGAG	5206
	GCAAGATCTAGATCAAG	520
	CTTGATCT A GATCTTGC	520
Male-sterile PI Zea mays	CTGCCCTTGAATCTGTTAGGGAGTGGAGAACGGAGTATGGGGCG CGGCAAGATCGAGATCTAGAGGATCGAGAACTCTACCAACCGGCA GGTGACCTTCTCCAAGCGCCGGGCCGG	
Lys9Term AAG-TAG	CCAGTCCGGCCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTA GAGTTCTCGATCCTCTAGATCTCGATCTTGCCGCGCCCCATACTC CGTTCTCCACTCCCTAACAGATTCAAGGGCAG	521

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TCGAGATC <u>T</u> AGAGGATC	5211
	GATCCTCTAGATCTCGA	5212
Male-sterile Pl Zea mays	AATCTGTTAGGGAGTGGAGAACGGAGTATGGGGCGCGCAAGAT CGAGATCAAGAGGATC <u>T</u> AGAACTCTACCAACCGGCAGGTGACCTT CTCCAAGCGCCGGGCCGG	5213
Glu12Term GAG-TAG	CCTTCTTGACCAGTCCGGCCCGGCGCTTGGAGAAGGTCACCTGC CGGTTGGTAGAGTTCTAGATCCTCTTGATCTCGATCTTGCCGCGC CCCATACTCCGTTCTCCACTCCCTAACAGATT	5214
	AGAGGATC <u>T</u> AGAACTCT	5215
	AGAGTTCT A GATCCTCT	5216
Male-sterile Pl Oryza sativa	TTGCTGCTAAGCTAGCTGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGA	5217
Lys5Term AAG-TAG	TGCGCTTGGAGAAGGTCACCTGGCGGTTGGTGGAGTTCTCGATCC TCTTGATCTCGATCTACCCGCGCCCCATCCCGCCTCCTCCTC CTCCTCCTTCCTCCAGCTAGCTTAGCAGCAA	5218
	GGCGCGG <u>T</u> AGATCGAG	5219
	CTCGATCT <u>A</u> CCCGCGCC	5220
Male-sterile Pl Oryza sativa	CTAAGCTAGCTGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGAGGA	5221
Glu7Term GAG-TAG	CGCTCCTGCGCTTGGAGAAGGTCACCTGGCGGTTGGTGGAGTTC TCGATCCTCTTGATCTAGATCTTCCCGCGCCCCATCCCGCCTCCT CCTCCTCCTCCTTCCTCCAGCTAGCTTAG	5222
	GGAAGATC <u>T</u> AGATCAAG	5223
	CTTGATCT <u>A</u> GATCTTCC	5224
Male-sterile Pl Oryza sativa	TAGCTGGAGGAGGAGGAGGAGGAGGAGGAGGAGGCGGGCG	5225
Lys9Term AAG-TAG	GGATCCCGCTCCTGCGCTTGGAGAAGGTCACCTGGCGGTTGGTG GAGTTCTCGATCCTCTAGATCTCCATCCCGCGCCCCCATCCCG CCTCCTCCTCCTCCTCCTCCAGCTA	5226
	TCGAGATC <u>T</u> AGAGGATC	5227
	GATCCTCT <u>A</u> GATCTCGA	5228

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile Pl	GAAGGAGGAGGAGGAGGAGGCGGGATGGGGCGCGGGAAGA TCGAGATCAAGAGGATC <u>T</u> AGAACTCCACCAACCGCCAGGTGACCT TCTCCAAGCGCAGGAGCGGGATCCTCAAGAAGG	5229
Oryza sativa Glu12Term GAG-TAG	CCTTCTTGAGGATCCCGCTCCTGCGCTTGGAGAAGGTCACCTGGC GGTTGGTGGAGTTCTAGATCCTCTTGATCTCGATCTTCCCGCGCC CCATCCCGCCTCCTCCTCCTCCTCCTTC	5230
	AGAGGATCTAGAACTCC	5231
	GGAGTTCT <u>A</u> GATCCTCT	5232

Example 7

Engineering plants for abiotic stress tolerance

Environmental stresses, such as drought, increased soil salinity, soil contamination with heavy metals, and extreme temperature, are major factors limiting plant growth and productivity. The worldwide loss in yield of three major cereal crops, rice, maize, and wheat due to water stress (drought) has been estimated to be over ten billion dollars annually and many currently marginal soils could be brought into cultivation if suitable plant varieties were available.

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Physiological and biochemical responses to high levels of ionic or nonionic solutes and decreased water potential have been studied in a variety of plants. It is known, for example, that increasing levels of alcohol dehydrogenase can confer enhances flooding resistance in plants. There are also several possible mechanisms to enhance plant salt tolerance. For example, one mechanism underlying the adaptation or tolerance of plants to osmotic stresses is the accumulation of compatible, low molecular weight osmolytes such as sugar alcohols, special amino acids, and glycinebetaine. Such accumulation can be engineered, for example, by removing feedback inhibition on 1-pyrroline-t-carboxylate synthetase, which results in accumulation of proline. Additionally, recent experiments suggest that altering the expression or activity of specific sodium or potassium transporters can confer enhanced salt tolerance.

Plant tolerance of contamination by heavy metals such as lead and aluminum in soils has also been investigated and one mechanism underlying tolerance is the production of dicarboxylic acids such as oxalate and citrate. In addition, individual genes involved in heavy metal sensitivity have been identified.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that confer stress tolerance in plants.

Table 17
Genome-Altering Oligos Conferring Stress Tolerance

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ NC
Salt Tolerance P5CS Arabidopsis thaliana	CGTCTTTTTGTGTGGTAGTTGGATGTGACGGTTGCTCAAATGCTT GTGACCGATAGCAGTGCTAGAGATAAGGATTTCAGGAAGCAACTT AGTGAAACTGTCAAAGCGATGCTGAGGATGA	52
Phe128Ala TTT-GCT	TCATCCTCAGCATCGCTTTGACAGTTTCACTAAGTTGCTTCCTGAA ATCCTTATCTCTAGCACTGCTATCGGTCACAAGCATTTGAGCAACC GTCACATCCAACTACCACACAAAAAGACG	52
	ATAGCAGT <u>GC</u> TAGAGAT	52
	ATCTCTA GC ACTGCTAT	52
Salt Tolerance P5CS 1 Brassica napus	GAGACTATGTTTGACCAGCTGGATGTGACGGCTGCTCAGCTGCTG GTGAATGACAGTAGTGCCAGAGACAAGGAGTTCAGGAAGCAACTT AATGAGACAGTGAAGTCCATGCTTGATTTGA	52
Phe128Ala TTC-GCC	TCAAATCAAGCATGGACTTCACTGTCTCATTAAGTTGCTTCCTGAA CTCCTTGTCTCTG <u>GC</u> ACTACTGTCATTCACCAGCAGCTGAGCAGC CGTCACATCCAGCTGGTCAAACATAGTCTC	5
	ACAGTAGTGCCAGAGAC	5
	GTCTCTGGCACTACTGT	5
Salt Tolerance P5CS 2	GAGACTATGTTTGACCAGATGGATGTGACGGTGGCTCAAATGCTG GTGACTGATAGCAGTGAAAGCTATGCTGAAAATGA	5
Brassica napus Phe129Ala TTC-GCC	TCATTTTCAGCATAGCTTTGACTGTCTCACTAAGTTGCTTCCTGAA ATCCTTATCTCTGACACTGCTATCAGTCACCAGCATTTGAGCCACC GTCACATCCATCTGGTCAAACATAGTCTC	
	ATAGCAGTGTCAGAGAT	
	ATCTCTGACACTGCTAT	
Salt Tolerance P5CS Oryza sativa	GATATGTTGAACCAACTGGATGTCTCGTCATCTCAACTTCTTG TCACCGACAGTGATGCTGAGAACCCAAAGTTCCGGGAGCAACTCA	_ _
Phe128Ala TTT-GCT	TAAGATCTAATAATGACTCAACAGTTTCAGTGAGTTGCTCCCGGAACTTTGGGTTCTCAGCATCACTGTCGGTGACAAGAAGTTGAGATGACAGAACATCCAGTTGGTTAAACAACATATC	
	ACAGTGATGCTGAGAAC	
	GTTCTCA GC ATCACTGT	

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Salt Tolerance P5CS Medicago sativa	GATATTTTGTTTAGTCAGCTGGATGTGACATCTGCTCAGCTTCTTG TTACTGACAATGATGCTAGAGAGCCAAGATTTTAGAAAGCAACTTTC TGAAACTGTGAGATCACTTCTAGCACTAA	5249
Phe128Ala TTT-GCT	TTAGTGCTAGAAGTGATCTCACAGTTTCAGAAAGTTGCTTTCTAAA ATCTTGGTCTCTA GC ATCATTGTCAGTAACAAGAAGCTGAGCAGAT GTCACATCCAGCTGACTAAACAAAATATC	5250
	ACAATGAT <u>GC</u> TAGAGAC	5251
	GTCTCTA GC ATCATTGT	5252
Salt Tolerance P5CS Actinidia deliciosa	GATACATTGTTTAGTCAGCTGGATGTGACATCAGCTCAGCTACTC GTTACTGATAATGATGCTAGGGATCCAGAATTCAGGAAGCAACTTA CTGAAACTGTAGAATCACTATTGAATTTGA	5253
Phe128Ala TTT-GCT	TCAAATTCAATAGTGATTCTACAGTTTCAGTAAGTTGCTTCCTGAAT TCTGGATCCCTA GC ATCATTATCAGTAACGAGTAGCTGAGCTGAT GTCACATCCAGCTGACTAAACAATGTATC	5254
	ATAATGAT GC TAGGGAT	5255
	ATCCCTA GC ATCATTAT	5256
Salt Tolerance P5CS Cichorium intybus	GACACACTCTTCAGTCAACTGGATGTGACATCAGCACAGCTTCTT GTAACAGATAATGAC GC CAGAAGTCCAGAATTTAGAAAACAACTTA CTGAAACAGTCGATTCTTTATTATCTTATA	5257
Phe122Ala TTC-GCC	TATAAGATAATAAAGAATCGACTGTTTCAGTAAGTTGTTTTCTAAAT TCTGGACTTCTG <u>GC</u> GTCATTATCTGTTACAAGAAGCTGTGCTGAT GTCACATCCAGTTGACTGAAGAGTGTGTC	5258
	ATAATGAC GC CAGAAGT	5259
	ACTTCTG GC GTCATTAT	5260
Salt Tolerance' P5CS Lycopersicon	GATTCTTTGTTCAGTCAGTTGGATGTGACATCAGCTCAGCTTCTGG TGACTGATAATGACGCTAGAGATCCAGATTTTAGGAGACAACTCAA TGACACAGTAAATTCGTTGCTTTCTCTAA	5261
esculentum Phe128Ala TTT-GCT	TTAGAGAAAGCAACGAATTTACTGTGTCATTGAGTTGTCTCCTAAA ATCTGGATCTCTAGCGTCATTATCAGTCACCAGAAGCTGAGCTGA TGTCACATCCAACTGACTGAACAAAGAATC	5262
	ATAATGAC <u>GC</u> TAGAGAT	5263
	ATCTCTA GC GTCATTAT	5264
Salt Tolerance P5CS Vigna unguiculata	GATACCATGTTCAGCCAGCTTGATGTGACTTCTTCCCAACTTCTTG TGAATGATGGATTT GC TAGGGATGCTGGCTTCAGAAAACAACTTTC GGACACAGTGAACGCGTTATTAGATTTAA	5265
Phe162Ala TTT-GCT	TTAAATCTAATAACGCGTTCACTGTGTCCGAAAGTTGTTTTCTGAA GCCAGCATCCCTAGCAAATCCATCATTCACAAGAAGTTGGGAAGA AGTCACATCAAGCTGGCTGAACATGGTATC	5266
	ATGGATTT GC TAGGGAT	5267

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	EQID NO:
	ATCCCTAGCAAAICCAI	5268
Salt Tolerance P5CS	GACACCTTGTTTAGTCAGTTGGATCTGACTGCTCAGCTGCTT GTGACGGACAACGACGCTAGAGATCCAAGTTTTAGAACACAACTA ACTGAAACAGTGTATCAGTTGTTGGATCTAA	5269
Phe125Ala	TTAGATCCAACAACTGATACACTGTTTCAGTTAGTTGTGTTCTAAAA	5270
TTT-GCT	GTCAGATCCAACTGACTAAACAAGGTGTC ACAACGACGCTAGAGAT	5271
,	ATCTCTA GC GTCGTTGT	5272
Salt Tolerance P5CS	GACACATTATTTAGCCAGCTGGATGTGACATCAGCTCAGCTTCTT GTGACTGATAATGATGCTAGGGATGAAGCTTTCCGAAATCAACTTA CTCAAACAGTGGATTCATTGTTAGCTTTGA	5273
Vitis vinifera Phe130Ala TTT-GCT	TCAAAGCTAACAATGAATCCACTGTTTGAGTAAGTTGATTTCGGAA	5274
	GTCACATCCAGCTGGCTAAATAATGTGTC ATAATGATGCTAGGGAT	527
	ATCCCTAGCATCATTAT	5270
Salt Tolerance P5CS	GATACGCTGTTCACTCAGCTCGATGTGACATCGGCTCAGCTTCTT GTGACGGATAACGAT GC TCGAGATAAGGATTTCAGGAAGCAGCTT	527
Vigna aconitifolia Phe129Ala TTT-GCT	ACTGAGACTGTGAAGTCGCTGTTGGCGCTGA TCAGCGCCAACAGCGACTTCACAGTCTCAGTAAGCTGCTTCCTGA AATCCTTATCTCGAGCATCGTTATCCGTCACAAGAAGCTGAGCCG ATGTCACATCGAGCTGAGTGAACAGCGTATC	527
	ATAACGATGCTCAGAT	527
	ATCTCGAGCATCGTTAT	528
Salt Tolerance HKT1	AGAGATGTTCTTAGTTCCAAAGAAATCTCACCTCTCACTTTCTCCG TCTTCACAACAGTTGTCACGTTTGCAAACTGCGGATTTGTCCCCAC	528
Arabidopsis thaliana Ser207Val TCC-GTC	TTTTGCGAAGATGATCATCTTTGGGVTT TTTTGCGAAGATGATCATCTTTCTCATTCGTGGGGACAAATCCGCA GTTTGCAAACGTGACAACTGTTGTGAAGACGGAGAAAGTGAGAGG TGAGATTTCTTTGGAACTAAGAACATCTCT	528
	CAACAGTT <u>GT</u> CACGTTT	52
	AAACGTGACAACTGTTG	52
Salt Tolerance HKT1	CGAATGAGAACATGATCATCTTTCGCAAAAACTCTGGTCTCATCTG GCTCCTAATCCCTCTAGTACTGATGGGAAACACTTTGTTCCCTTGC TTCTTGGTTTTGCTCATATGGGGACTTTA	
Arabidopsis thaliana Gln237Leu CAA-CTA	TAAAGTCCCCATATGAGCAAAACCAAGAAGCAAGGGAACAAAGTG TTTCCCATCAGTACTAGAGGGATTAGGAGCCAGATGAGACCAGAG TTTTTGCGAAAGATGATCATGTTCTCATTCG	52

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	AATCCCTC <u>T</u> AGTACTGA	5287
	TCAGTACT <u>A</u> GAGGGATT	5288
Salt Tolerance	AGTCTCTAGAAGGAATGAGTTCGTACGAGAAGTTGGTTGG	5289
HKT1	TGTTTCAAGTGGTGAGTTCGCGACACACCGGAGAAACTATAGTAG	
Arabidopsis thaliana	ACCTCTCTACACTTTCCCCAGCTATCTTGGT	5000
Asn332Ser AAT-AGT	ACCAAGATAGCTGGGGAAAGTGTAGAAAGAAGAAAGAAAG	5290
AAI-AGI	CCGGTGTGTCGCGAACTCACCACTTGAAACAACGATCCAACCAA	
	AGTGGTGAGACTCATTCCTTCTAGAGACT	5291
	GTCGCGAACTCACCACT	5292
0.47.1		
Salt Tolerance HKT1	AGAGATGTGCTAAAGAAGAAAGGTCTCAAAATGGTGACCTTTTCC GTCTTCACCACCGTGGTGACCTTTGCCAGTTGTGGGTTTGTCCCG	5293
Eucalyptus	ACCAATGAAAACATGATTATCTTCAGCAAAA	
camaldulensis	TTTTGCTGAAGATAATCATGTTTTCATTGGTCGGGACAAACCCACA	5294
Ser256Val	ACTGGCAAAGGTCACCACGGTGGTGAAGACGGAAAAGGTCACCA	020.
TCG-GTG	TTTTGAGACCTTTCTTTAGCACATCTCT	
,	CCACCGTG <u>GT</u> GACCTTT	5295
·	AAAGGTC <u>AC</u> CACGGTGG	5296
Salt Tolerance	CCAATGAAAACATGATTATCTTCAGCAAAAACTCTGGCCTCCTCCT	5297
HKT1	GATTCTCATCCCTC <u>T</u> GGCCCTTCTTGGGAACATGCTGTTCCCATC	
Eucalyptus	GAGCCTACGTTTGACGCTTTGGCTCATCGG	
camaldulensis	CCGATGAGCCAAAGCGTCAAACGTAGGCTCGATGGGAACAGCAT	5298
Gln286Leu	GTTCCCAAGAAGGCCC <u>A</u> GAGGGATGAGAATCAGGAGGAGGCCA	
CAG-CTG	GAGTTTTTGCTGAAGATAATCATGTTTTCATTGG CATCCCTCTGGCCCTTC	5299
	GAAGGCC <u>A</u> GAGGGATG	5300
Salt Tolerance	AATCGTTGAATGGACTAAGCTCCTGTGAGAAAATCGTGGGCGCGC	5301
HKT1	TGTTTCAGTGCGTGA <u>G</u> CAGCAGACATACCGGCGAGACGGTCGTC	
Eucalyptus	GATCTGTCCACAGTTGCTCCCGCCATCTTGGT	5000
camaldulensis Asn381Ser	ACCAAGATGGCGGGAGCAACTGTGGACAGATCGACGACCACCA	5302
AS0301Ser AAC-AGC	GCCGGTATGTCTGCTGCTCACGCACTGAAACAGCGCGCCCACGA TTTTCTCACAGGAGCTTAGTCCATTCAACGATT	
7770-700	GTGCGTGAGCAGCAGAC	5303
	GTCTGCTGCTCACGCAC	5304

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Salt Tolerance	AAAGCTCCACTGAAGAAGAAAGGGATCAACATTGCACTCTCTCAT TCTCGGTCACGGTCGTCCCGTTCTCAACAACAACA	5305
O <i>ryza sativa</i> Ser238Val FCC-GTC	ACAAATGAGAACATGGCAATCTTCTCCAAGA TCTTGGAGAAGATTGCCATGTTCTCATTTGTCGGCACGAGCCCCA CATTCGCAAACGAGACGACGACCGTGACCGAGAATGAGAAGAGTGCA ATGTTGATCCCTTTCTTCTTCAGTGGAGCTTT	5306
	TCACGGTCGTTTTTTCAGTGGAGGTTTTTTTCAGTGGAGGTTTTTTTT	5307
	AAACGAGACGACCGTGA	5308
Salt Tolerance HKT1	CAAATGAGAACATGGCAATCTTCTCCAAGAACCCGGGCCTCCTCC TCCTGTTCATCGGCCTGATTCTTGCAGGCAATACACTTTACCCTCT CTTCCTAAGGCTATTGATATGGTTCCTGGG	5309
Oryza sativa Gln268Leu CAG-CTG	CCCAGGAACCATATCATAGCCTTAGGAAGAGAGGGGTAAAGTGTA TTGCCTGCAAGAATCAGGCCGATGAACAGGAGGAGGAGGCCCGG GTTCTTGGAGAAGATTGCCATGTTCTCATTTG	5310
	CATCGGCC <u>T</u> GATTCTTG	5311
	CAAGAATCAGGCCGATG	5312
Salt Tolerance HKT1	CAGTCTTTGATGGACTCAGCTCTTACCAGAAGATTATCAATGCATT GTTCATGGCAGTGAGCGCAAGGCACTCGGGGGAGAACTCCATCG ACTGCTCACTCATCGCCCCTGCTGTTCTAGT	5313
Oryza sativa Asn363Ser AAC-AGC	ACTAGAACAGCAGGGGCGATGAGTGAGCAGTCGATGGAGTTCTC CCCCGAGTGCCTTGCGCTCACTGCCATGAACAATGCATTGATAAT	5314
	CTTCTGGTAAGAGCTGAGTCCATCAAAGACTG GGCAGTGA <u>G</u> CGCAAGGC	5318
	GCCTTGCGCTCACTGCC	5310
Salt Tolerance HKT1	GTGCCCACTGAACAAGAAAGGGATCAACATCGTGCTCTTCTCAC TATCAGTCACCGTTGTCTCCTGTGCGAATGCAGGACTCGTGCCCA CAAATGAGAACATGGTCATCTTCTCAAAGAA	531
Triticum aestivum Ala240Val GCC-GTC	TTCTTTGAGAAGATGACCATGTTCTCATTTGTGGGCACGAGTCCT GCATTCGCACAGGAGACAACGGTGACTGATAGTGAGAAGAGCAC GATGTTGATCCCTTTCTTGTTCAGTGGGGCAC	531
	CACCGTTGTCTCCTGTG	531
	CACAGGAGACAACGGTG	532
Salt Tolerance HKT1	CAAATGAGAACATGGTCATCTTCTCAAAGAATTCAGGCCTCTTGTT GCTGCTGAGTGGCCTGATGCTCGCAGGCAATACATTGTTCCCTCT CTTCCTGAGGCTACTGGTGTGGTTCCTGGG	532
Triticum aestivum Gln270Leu CAG-CTG	CCCAGGAACCACCAGTAGCCTCAGGAAGAGAGGGAACAATGT ATTGCCTGCGAGCATCAGGCCACTCAGCAGCAACAAGAGGCCTG AATTCTTTGAGAAGATGACCATGTTCTCATTTG	532
	GAGTGCCTGATGCTCG	532

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CGAGCATC <u>A</u> GGCCACTC	5324
Salt Tolerance HKT1 <i>Triticum aestivum</i>	CAGTCTTTGATGGGCTCAGCTCTTATCAGAAGACTGTCAATGCATT CTTCATGGTGGTGAGTGCGAGGCACTCAGGGGAGAATTCCATCG ACTGCTCGCTCATGTCCCCTGCCATTATAGT	5325
Asn365Ser AAT-AGT	ACTATAATGGCAGGGGACATGAGCGAGCAGTCGATGGAATTCTCC CCTGAGTGCCTCGCACCACCATGAAGAATGCATTGACAGTC TTCTGATAAGAGCTGAGCCCATCAAAGACTG	5326
	GGTGGTGAGGC	5327
	GCCTCGCA <u>C</u> TCACCACC	5328
Freezing Tolerance proline oxidase precursor	TTTTTTTGTTTTCGTTTTCAAAAACAAAATCTTTGAATTTTATGGCA ACCCGTCTTCTCTGAACAAACTTTATCCGGCGATCTTACCGTTTAC CCGCTTTTAGCCCGGTGGGTCCTCCCA	5329
Arabidopsis thaliana Arg7Term CGA-TGA	TGGGAGGACCCACCGGGCTAAAAGCGGTAAACGGTAAGATCGC CGGATAAAGTTTGTTCAGAGAAGACGGGTTGCCATAAAATTCAAA GATTTTGTTTTTGAAAAACGAAAAAAAAAA	5330
	GTCTTCTCTGAACAAAC	5331
	GTTTGTTC <u>A</u> GAGAAGAC	5332
Freezing Tolerance proline oxidase precursor	TCAAAAACAAAATCTTTGAATTTTATGGCAACCCGTCTTCTCAGAA CAAACTTTATCCGGTGATCTTACCGTTTACCCGCTTTTAGCCCGGT GGGTCCTCCCACCGTGACTGCTTCCACCG	5333
Arabidopsis thaliana Arg13Term CGA-TGA	CGGTGGAAGCAGTCACGGTGGGAGGACCCACCGGGCTAAAAGC GGGTAAACGGTAAGATCACCGGATAAAGTTTGTTCTGAGAAGACG GGTTGCCATAAAATTCAAAGATTTTGTTTTTGA	5334
	TTATCCGG <u>T</u> GATCTTAC	5335
	GTAAGATC <u>A</u> CCGGATAA	5336
Freezing Tolerance proline oxidase precursor	AAAATCTTTGAATTTTATGGCAACCCGTCTTCTCCGAACAACTTTA TCCGGCGATCTTAGCCGGTTTTAGCCCGGTGGGTCCTC CCACCGTGACTGCTTCCACCGCCGTCGTC	5337
<i>Arabidopsis thaliana</i> Tyr15Term TAC-TAG	GACGACGCGGTGGAAGCAGTCACGGTGGGAGGACCCACCGGG CTAAAAGCGGGTAAACGCTAAGATCGCCGGATAAAGTTTGTTCGG AGAAGACGGGTTGCCATAAAATTCAAAGATTTT	5338
	CGATCTTA <u>G</u> CGTTTACC	5339
	GGTAAACG C TAAGATCG	5340

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	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Freezing Tolerance proline oxidase	CTTTGAATTITATGGCAACCCGTCTTCTCCGAACAAACTTTATCCG GCGATCTTACCGTT <u>A</u> ACCCGCTTTTAGCCCGGTGGGTCCTCCCAC CGTGACTGCTTCCACCGCCGTCGTCCCGGA	5341
5	Archidoncic thaliana	TCCGGGACGACGGTGGAAGCAGTCACGGTGGGAGGACCCA CCGGGCTAAAAGCGGGTTAACGGTAAGATCGCCGGATAAAGTTTG TTCGGAGAAGACGGGTTGCCATAAAATTCAAAG	5342
	11A-1AA	TTACCGTT <u>A</u> ACCCGCTT	5343
		AAGCGGGT <u>T</u> AACGGTAA	5344
	Freezing Tolerance proline oxidase	CCGGTGGTCCTCCCACCGTGACTGCTTCCACCGCCGTCGTCCC GGAGATTCTCTCTTTTGACAACAAGCACCGGAACCACCTCTTCA CCACCCAAAACCCACCGAGCAATCTCACGATG	5345
10	precursor Arabidopsis thaliana Gly42Term GGA-TGA	CATCGTGAGATTGCTCGGTGGGTTTTGGGTGGTGAAGAGGTGGT TCCGGTGCTTGTTGTCAAAAGGAGAGAATCTCCGGGACGACGGC GGTGGAAGCAGTCACGGTGGGAGGACCCACCGG	5346
	GGA-1GA	TCTCCTTT <u>T</u> GACAACAA	5347
		TTGTTGTC <u>A</u> AAAGGAGA	5348
15	Lead Tolerance cyclic nucleotide- regulated ion channel	ACATGAAGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCA AACTATGAATTTCTGACAAGAGAAGTTTGTAAGGTCAGTGTTCCAG ATTTGTCTCATTGAATTCTAAGTCGTGA	5349
15	Arabidopsis thaliana Arg4Term CGA-TGA	TCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGACCTTAC AAACTTCTCTTGTCAGAAATTCATAGTTTGAGACTAATAAGATTCAA TACAAACAGAGATTTCACTGCTTCATGT	5350
	CGA-1GA	TGAATTTC <u>T</u> GACAAGAG	5351
		CTCTTGTC <u>A</u> GAAATTCA	5352
20	Lead Tolerance cyclic nucleotide-	TGAAGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAAC TATGAATTTCCGATAAGAGAAGTTTGTAAGGTCAGTGTTCCAGATT TGTCTCATTGAATTCTAAGTCGTGAAGC	5353
	regulated ion channel Arabidopsis thaliana Gln5Term	GCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGACCT TACAAACTTCTCTTATCGGAAATTCATAGTTTGAGACTAATAAGATT	5354
	CAA-TAA	CAATACAAACAGAGATTTCACTGCTTCA ATTTCCGA <u>T</u> AAGAGAAG	5355
		CTTCTCTTATCGGAAAT	5356
25	Lead Tolerance cyclic nucleotide-	AGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAACTAT GAATTTCCGACAATAGAAGTTTGTAAGGTCAGTGTTCCAGATTTGT CTCATTGAATTCTAAGTCGTGAAGCTTA	5357
30	regulated ion channel Arabidopsis thaliana Glu6Term GAG-TAG	TAAGCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGA CCTTACAAACTTCTATTGTCGGAAATTCATAGTTTGAGACTAATAA GATTCAATACAAACAGAGATTTCACTGCT	5358

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	TCCGACAA <u>T</u> AGAAGTTT	5359
	AAACTTCT <u>A</u> TTGTCGGA	5360
Lead Tolerance cyclic nucleotide- regulated ion channel Arabidopsis thaliana Lys7Term AAG-TAG	AGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAACTATGAA TTTCCGACAAGAGTAGTTTGTAAGGTCAGTGTTCCAGATTTGTCTC ATTGAATTCTAAGTCGTGAAGCTTAATT	5361
	AATTAAGCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACAC TGACCTTACAAACT <u>A</u> CTCTTGTCGGAAATTCATAGTTTGAGACTAA TAAGATTCAATACAAACAGAGATTTCACT	5362
	GACAAGAG <u>T</u> AGTTTGTA	5363
	TACAAACT <u>A</u> CTCTTGTC	5364
Lead Tolerance cyclic nucleotide- regulated ion channel Arabidopsis thaliana Gln12Term CAA-TAA	CATTGAATTCTAAGTCGTGAAGCTTAATTCGATTCTTCACTTTC TCGGATCAGGTTTTAAGATTGGAAGTCGGATAAGACTTCCTCCGA CGTGGAATATTCCGGTAAAAACGAGATTC	5365
	GAATCTCGTTTTTACCGGAATATTCCACGTCGGAGGAAGTCTTATC CGACTTCCAATCTT <u>A</u> AAACCTGATCCGAGAAAGTGAAGAAGAATC GAATTAAGCTTCACGACTTAGAATTCAATG	5366
	TCAGGTTT <u>T</u> AAGATTGG	5367
	CCAATCTT <u>A</u> AAACCTGA	5368
Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) Nicotiana Tabacum GIn5Term CAA-TAA	TGGAAGTCAATCCCCCACGTTGAGCAGGTTGATGCATTGGCTAAA GTTATGAATCACCGC <u>T</u> AAGACGAGTTTGTGAGGTTTCAGGATTGG AAATCAGAGAGAAGCTCTGAGGGAAATTTTC	5369
	GAAAATTTCCCTCAGAGCTTCTCTCTGATTTCCAATCCTGAAACCT CACAAACTCGTCTTAGCGGTGATTCATAACTTTAGCCAATGCATCA ACCTGCTCAACGTGGGGGGATTGACTTCCA	5370
	ATCACCGCTAAGACGAG	5371
	CTCGTCTT <u>A</u> GCGGTGAT	5372
Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) Nicotiana Tabacum Gly7Term GAG-TAG	TCAATCCCCCACGTTGAGCAGGTTGATGCATTGGCTAAAGTTATG AATCACCGCCAAGAC <u>T</u> AGTTTGTGAGGTTTCAGGATTGGAAATCA GAGAGAAGCTCTGAGGGAAATTTTCATGCTA	5373
	TAGCATGAAAATTTCCCTCAGAGCTTCTCTCTGATTTCCAATCCTG AAACCTCACAAACTAGTCTTGGCGGTGATTCATAACTTTAGCCAAT GCATCAACCTGCTCAACGTGGGGGGATTGA	5374
	GCCAAGAC <u>T</u> AGTTTGTG	5375
	CACAAACT <u>A</u> GTCTTGGC	5376

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Lead Tolerance cyclic nucleotide- gated calmodulin-	GAGCAGGTTGATGCATTGGCTAAAGTTATGAATCACCGCCAAGAC GAGTTTGTGAGGTTT <u>T</u> AGGATTGGAAATCAGAGAGAAGCTCTGAG GGAAATTTTCATGCTAAAGGTGGAGTCCACC	5377
	binding ion channel (CBP4) Nicotiana Tabacum	GGTGGACTCCACCTTTAGCATGAAAATTTCCCTCAGAGCTTCTCTC TGATTTCCAATCCTAAAACCTCACAAACTCGTCTTGGCGGTGATTC ATAACTTTAGCCAATGCATCAACCTGCTC	5378
	Gln12Term	TGAGGTTT <u>T</u> AGGATTGG	5379
	CAG-TAG	CCAATCCT <u>A</u> AAACCTCA	5380
	Lead Tolerance cyclic nucleotide- gated calmodulin-	TGATGCATTGGCTAAAGTTATGAATCACCGCCAAGACGAGTTTGT GAGGTTTCAGGATTG <u>T</u> AAATCAGAGAGAAGCTCTGAGGGAAATTT TCATGCTAAAGGTGGAGTCCACCGAAGTAAA	5381
	binding ion channel (CBP4)	TITACTTCGGTGGACTCCACCTTTAGCATGAAAATTTCCCTCAGAG CTTCTCTCTGATTT <u>A</u> CAATCCTGAAACCTCACAAACTCGTCTTGGC GGTGATTCATAACTTTAGCCAATGCATCA	5382
	Trp14Term	CAGGATTG <u>T</u> AAATCAGA	5383
	TGG-TGA	TCTGATIT <u>A</u> CAATCCTG	5384
	Lead Tolerance cyclic nucleotide- gated calmodulin-	GATGCATTGGCTAAAGTTATGAATCACCGCCAAGACGAGTTTGTG AGGTTTCAGGATTGGTAATCAGAGAGAAGCTCTGAGGGAAATTTT CATGCTAAAGGTGGAGTCCACCGAAGTAAAG	5385
	binding ion channel (CBP4) Nicotiana Tabacum	CTITACTTCGGTGGACTCCACCTITAGCATGAAAATTTCCCTCAGA GCTTCTCTCTGATTACCAATCCTGAAACCTCACAAACTCGTCTTGG CGGTGATTCATAACTTTAGCCAATGCATC	5386
	Lys15Term	AGGATTGGTAATCAGAG	5387
	AAA-TAA	CTCTGATT <u>A</u> CCAATCCT	5388
	Lead Tolerance calmodulin binding	CTTGAAGAATTGATCTACCACTCTTAGCTGCTAACTGTTCGCCTGG TGGAGATAATGATGTAAAGAGAGGACAGATATGTTAGATTTCAGGA CTGCAAATCAGAGCAATCTGTTATCTCAG	5389
	transport protein Hordeum vulgare Glu2Term	CTGAGATAACAGATTGCTCTGATTTGCAGTCCTGAAATCTAACATA TCTGTCCTCTCTTTACATCATTATCTCCACCAGGCGAACAGTTAGC AGCTAAGAGTGGTAGATCAATTCTTCAAG	5390
)	GAA-TAA	TAATGATG <u>T</u> AAAGAGAG	539
		CTCTCTTTACATCATTA	5392

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Lead Tolerance calmodulin binding transport protein	GAAGAATTGATCTACCACTCTTAGCTGCTAACTGTTCGCCTGGTG GAGATAATGATGGAA <u>T</u> GAGAGGACAGATATGTTAGATTTCAGGAC TGCAAATCAGAGCAATCTGTTATCTCAGAGA	5393
5	Hordeum vulgare Arg3Term AGA-TGA	TCTCTGAGATAACAGATTGCTCTGATTTGCAGTCCTGAAATCTAAC ATATCTGTCCTCTCATCATCATCATCATCACCACCAGGCGAACAGTT AGCAGCTAAGAGTGGTAGATCAATTCTTC	5394
		TGATGGAA <u>T</u> GAGAGGAC	5395
		GTCCTCTCATTCCATCA	5396
	Lead Tolerance calmodulin binding transport protein	GAATTGATCTACCACTCTTAGCTGCTAACTGTTCGCCTGGTGGAG ATAATGATGGAAAGATAGGACAGATATGTTAGATTTCAGGACTGCA AATCAGAGCAATCTGTTATCTCAGAGAACG	5397
10	Hordeum vulgare Glu4Term GAG-TAG	CGTTCTCTGAGATAACAGATTGCTCTGATTTGCAGTCCTGAAATCT AACATATCTGTCCTATCTTTCCATCATTATCTCCACCAGGCGAACA GTTAGCAGCTAAGAGTGGTAGATCAATTC	5398
		TGGAAAGA <u>T</u> AGGACAGA	5399
		TCTGTCCT <u>A</u> TCTTTCCA	5400
15	Lead Tolerance calmodulin binding transport protein	ATCTACCACTCTTAGCTGCTAACTGTTCGCCTGGTGGAGATAATG ATGGAAAGAGGGACTGATATGTTAGATTTCAGGACTGCAAATCA GAGCAATCTGTTATCTCAGAGAACGCAGTTT	5401
	Hordeum vulgare Arg6Term AGA-TGA	AAACTGCGTTCTCTGAGATAACAGATTGCTCTGATTTGCAGTCCTG AAATCTAACATATCAGTCCTCTCTTTCCATCATTATCTCCACCAGG CGAACAGTTAGCAGCTAAGAGTGGTAGAT	5402
		GAGAGGAC <u>T</u> GATATGTT	5403
		AACATATC <u>A</u> GTCCTCTC	5404
20	Lead Tolerance calmodulin binding transport protein	CCACTCTTAGCTGCTAACTGTTCGCCTGGTGGAGATAATGATGGA AAGAGAGGACAGATAGGTTAGATTTCAGGACTGCAAATCAGAGCA ATCTGTTATCTCAGAGAACGCAGTTTCACCA	5405
	Hordeum vulgare Tyr7Term TAT-TAG	TGGTGAAACTGCGTTCTCTGAGATAACAGATTGCTCTGATTTGCAG TCCTGAAATCTAACCTATCTGTCCTCTTTTCCATCATTATCTCCAC CAGGCGAACAGTTAGCAGCTAAGAGTGG	5406
		GACAGATA G GTTAGATT	5407
		AATCTAAC <u>C</u> TATCTGTC	5408
25	2,4-DB resistance 3-ketoacyl-CoA thiolase	ATCCTTCTCTGAGAAAAAACAACAGATCCGAATTTTATCTTTAATCA GCCGGAAAAAATGTAGAAAGCGATCGAGAGACAACGCGTTCTTCT TGAGCATCTCCGACCTTCTTCTTCTTCTT	5409
30	Arabidopsis thaliana Glu2Term GAG-TAG	AAGAAGAAGAAGAAGGTCGGAGATGCTCAAGAAGAACGCGTTGTC TCTCGATCGCTTTCTACATTTTTTCCGGCTGATTAAAGATAAAATTC GGATCTGTTTTTTTCTCAGAGAAGGAT	5410

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Atteration	AAAAAATGTAGAAAGCG	5411
	CGCTTTCT <u>A</u> CATTTTT	5412
2,4-DB resistance 3-ketoacyl-CoA	CTTCTCTGAGAAAAACAACAGATCCGAATTTTATCTTTAATCAGC CGGAAAAAATGGAGTAAGCGATCGAGAGACAACGCGTTCTTCTTG	5413
hiolase Arabidopsis thaliana Lys3Term	AGCATCTCCGACCTTCTTCTTCTTCGC GCGAAGAAGAAGAAGAAGGTCGGAGATGCTCAAGAAGAACGCGT TGTCTCTCGATCGCTTACTCCATTTTTTCCGGCTGATTAAAGATAA AATTCGGATCTGTTTTTTCTCAGAGAAG	5414
AAA-TAA	AAATGGAGTAAGCGATC	5415
	GATCGCTTACTCCATTT	5416
2,4-DB resistance 3-ketoacyl-CoA	GAAAAAACAACAGATCCGAATTTTATCTTTAATCAGCCGGAAAAAA TGGAGAAAGCGATCTAGAGACAACGCGTTCTTCTTGAGCATCTCC GACCTTCTTCTTCTTCTTCGCACAATTACG	5417
thiolase Arabidopsis thaliana Glu6Term	CGTAATTGTGCGAAGAAGAAGAAGAAGGTCGGAGATGCTCAAGAA GAACGCGTTGTCTCTAGATCGCTTTCTCCATTTTTTCCGGCTGATT AAAGATAAAATTCGGATCTGTTGTTTTTC	5418
GAG-TAG	AAGCGATCTAGAGACAA	5419
	TTGTCTCTAGATCGCTT	5420
2,4-DB resistance 3-ketoacyl-CoA	AAAACAACAGATCCGAATTTTATCTTTAATCAGCCGGAAAAAATGG AGAAAGCGATCGAGTGACAACGCGTTCTTCTTGAGCATCTCCGAC CTTCTTCTTCTTCTCGCACAATTACGAGG	542
thiolase Arabidopsis thaliana Arg7Term AGA-TGA	CCTCGTAATTGTGCGAAGAAGAAGAAGAAGAAGGTCGGAGATGCTCAA GAAGAACGCGTTGTCACTCGATCGCTTTCTCCATTTTTCCGGCT GATTAAAGATAAAATTCGGATCTGTTGTTTT	542
AGA-1GA	CGATCGAGTGACAACGC	542
	GCGTTGTCACTCGATCG	542
2,4-DB resistance 3-ketoacyl-CoA	ACAACAGATCCGAATTITATCTTTAATCAGCCGGAAAAAATGGAGA AAGCGATCGAGAGATAACGCGTTCTTCTTGAGCATCTCCGACCTT CTTCTTCTTCTCGCACAATTACGAGGCTT	542
thiolase Arabidopsis thaliana Gln8Term	AAGCCTCGTAATTGTGCGAAGAAGAAGAAGAAGAAGGTCGGAGATGCT CAAGAAGAACGCGTTATCTCTCGATCGCTTTCTCCATTTTTTCCGG CTGATTAAAGATAAAATTCGGATCTGTTGT	542
CAA-TAA	TCGAGAGATAACGCGTT	542
	AACGCGTTATCTCTCGA	542

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
2,4-DB resistance glyoxysomal beta- ketoacyol-thiolase	GAGAGACAAAGAGTTCTTCTTGAACATCTCCGTCCTTCTTCTTCTT CCTCTCACAGCTTTTAAGGCTCTCTCTCTCTCAGCTTGCTT	5429
precursor <i>Brassica napus</i> Glu26Term	AGGTCCTCTGATACGCAGCACTGTCCCCAGCCAAGCAAGC	5430
GAA-TAA	ACAGCTTT <u>T</u> AAGGCTCT	5431
	AGAGCCTT <u>A</u> AAAGCTGT	5432
2,4-DB resistance glyoxysomal beta- ketoacyol-thiolase	TTGAACATCTCCGTCCTTCTTCTTCTTCCTCACAGCTTTGAAGG CTCTCTCTCTGCTTGAGCTTGCTTGGCTGGGGACAGTGCTGCGTA TCAGAGGACCTCTCTCTATGGAGATGATGT	5433
precursor Brassica napus Ser32Term	ACATCATCTCCATAGAGAGAGGTCCTCTGATACGCAGCACTGTCC CCAGCCAAGCAAGCTCAAGCAGAGAGAGAGCCTTCAAAGCTGTG AGAGGAAGAAGAAGAAGGACGGAGATGTTCAA	5434
TCA-TGA	CTCTGCTT <u>G</u> AGCTTGCT	5435
	AGCAAGCT <u>C</u> AAGCAGAG	5436
2,4-DB resistance glyoxysomal beta- ketoacyol-thiolase	TCTCCGTCCTTCTTCTTCCTCTCACAGCTTTGAAGGCTCTCTC TCTGCTTCAGCTTGATTGGCTGGGGACAGTGCTGCGTATCAGAG GACCTCTCTCTATGGAGATGATGTAGTCATT	5437
precursor Brassica napus Cys34Term	AATGACTACATCTCCATAGAGAGAGGTCCTCTGATACGCAGC ACTGTCCCCAGCCAA <u>T</u> CAAGCTGAAGCAGAGAGAGAGCCTTCAAA GCTGTGAGAGGAAGAAGAAGAAGGACGGAGA	5438
TGC-TGA	TCAGCTTG <u>A</u> TTGGCTGG	5439
	CCAGCCAA <u>T</u> CAAGCTGA	5440
2,4-DB resistance glyoxysomal beta- ketoacyol-thiolase	TCCGTCCTTCTTCTTCCTCTCACAGCTTTGAAGGCTCTCTCT	5441
precursor Brassica napus Leu35Term	ACAATGACTACATCATCTCCATAGAGAGAGGGTCCTCTGATACGCA GCACTGTCCCCAGCC <u>T</u> AGCAAGCTGAAGCAGAGAGAGAGCCTTC AAAGCTGTGAGAGGAAGAAGAAGAAGAAGGACGGA	5442
TTG-TAG	AGCTTGCT <u>A</u> GGCTGGGG	5443
	CCCCAGCCTAGCAAGCT	5444

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	2,4-DB resistance glyoxysomal beta- ketoacyol-thiolase	TCACAGCTTTGAAGGCTCTCTCTCTCTCTCAGCTTGCTTG	5445
5	precursor Brassica napus Tyr42Term	TAGTGCAGTCCTATGTGCCGCAACAATGACTACATCATCTCCATA GAGAGAGGTCCTCTGCTACGCAGCACTGTCCCCAGCCAAGCAAG	5446
	TAT-TAG	GCTGCGTA <u>G</u> CAGAGGAC	5447
		GTCCTCTGCTACGCAGC	5448
10	2,4-DB resistance 3-ketoacyl-CoA thiolase B	CAACAGACAGCAAGTGTTGCTCCAGCATCTCCGCCCTTCTAATTC TTCTTCTCACAATTAGGAGTCCGCTCTTGCCGCATCAGTATGTGCT GCAGGGGATAGCGCCGCATATCATAGGGCT	5449
10	Mangifera indica Tyr25Term TAC-TAG	AGCCCTATGATATGCGGCGCTATCCCCTGCAGCACATACTGATGC GGCAAGAGCGGACTCCTAATTGTGAGAAGAAGAATTAGAAGGGC GGAGATGCTGGAGCAACACTTGCTGTCTGTTG	5450
	170-170	CACAATTA <u>G</u> GAGTCCGC	5451
		GCGGACTC <u>C</u> TAATTGTG	5452
15	2,4-DB resistance 3-ketoacyol-CoA thiolase B	AACAGACAGCAAGTGTTGCTCCAGCATCTCCGCCCTTCTAATTCTT CTTCTCACAATTACTAGTCCGCTCTTGCCGCATCAGTATGTGCTG CAGGGGATAGCGCCGCATATCATAGGGCTT	5453
	Magnifera indica Glu26Term	AAGCCCTATGATATGCGGCGCTATCCCCTGCAGCACATACTGATG CGGCAAGAGCGGACTAGTAATTGTGAGAAGAAGAATTAGAAGGG CGGAGATGCTGGAGCAACACTTGCTGTCTGTT	5454
	GAG-TAG	ACAATTAC <u>T</u> AGTCCGCT	5455
		AGCGGACT A GTAATTGT	5456
20	2,4-DB resistance 3-ketoacy\ol-CoA thiolase B	TCCAGCATCTCCGCCCTTCTAATTCTTCTCACAATTACGAGTC CGCTCTTGCCGCATGAGTATGTGCTGCAGGGGATAGCGCCGCAT ATCATAGGGCTTCTGTTTATGGAGACGATGT	5457
0.E	Mangifera indica Ser32Term	ACATCGTCTCCATAAACAGAAGCCCTATGATATGCGGCGCTATCC CCTGCAGCACATACTCATGCGGCAAGAGCGGACTCGTAATTGTGA GAAGAAGAATTAGAAGGGCGGAGATGCTGGA	5458
25	TCA-TGA	TGCCGCATGAGTATGTG	5459
		CACATACT <u>C</u> ATGCGGCA	5460

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
2,4-DB resistance 3-ketoacyl-CoA thiolase B	TCTCCGCCCTTCTAATTCTTCTCACAATTACGAGTCCGCTCTT GCCGCATCAGTATGAGCTGCAGGGGATAGCGCCGCATATCATAG GGCTTCTGTTTATGGAGACGATGTGGTGATT	5461
Mangifera indica Cys34Term TGT-TGA	AATCACCACATCGTCTCCATAAACAGAAGCCCTATGATATGCGGC GCTATCCCCTGCAGC <u>T</u> CATACTGATGCGGCAAGAGCGGACTCGT AATTGTGAGAAGAAGAATTAGAAGGGCGGAGA	5462
	TCAGTATG <u>A</u> GCTGCAGG	5463
	CCTGCAGC <u>T</u> CATACTGA	5464
2,4-DB resistance 3-ketoacyl-CoA thiolase B	TCACAATTACGAGTCCGCTCTTGCCGCATCAGTATGTGCTGCAGG GGATAGCGCCGCATAGCATA	5465
Mangifera indica Tyr42Term TAT-TAG	AAGTGCAGTACGATGAGCTGCCACAATCACCACATCGTCTCCATA AACAGAAGCCCTATGCTATG	5466
	GCCGCATA G CATAGGGC	5467
	GCCCTATG <u>C</u> TATGCGGC	5468
2,4-DB resistance 3-ketoacyl-CoA thiolase	GAAGGCGATCAACAGGCAGAGCATTTTGCTACATCATCTCCGGCC TTCTTCTTCCGCTTAGACAAATGAATCTTCGCTCTCTGCATCGGTT TGTGCAGCTGGGGATAGTGCTTCGTATCAA	5469
Cucumis sativus Tyr22Term TAC-TAG	TTGATACGAAGCACTATCCCCAGCTGCACAAACCGATGCAGAGAGCGAAGATTCATTTGTCTAAGCGGAAGAAGAAGCCCGGAGATGATGTAGCAAAATGCTCTGCCTGTTGATCGCCTTC	5470
	TCCGCTTA <u>G</u> ACAAATGA	5471
	TCATTTGT <u>C</u> TAAGCGGA	5472
2,4-DB resistance 3-ketoacyl-CoA thiolase	ATCAACAGGCAGAGCATTTTGCTACATCATCTCCGGCCTTCTTCTT CCGCTTACACAAATTAATCTTCGCTCTCTGCATCGGTTTGTGCAGC TGGGGATAGTGCTTCGTATCAAAGGACAT	5473
Cucumis sativus Glu25Term GAA-TAA	ATGTCCTTTGATACGAAGCACTATCCCCAGCTGCACAAACCGATG CAGAGAGCGAAGATTAATTTGTGTAAGCGGAAGAAGAAGGCCGG AGATGATGTAGCAAAATGCTCTGCCTGTTGAT	5474
•	ACACAAAT <u>T</u> AATCTTCG	5475
	CGAAGATT <u>A</u> ATTTGTGT	5476

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
2,4-DB resistance 3-ketoacyl-CoA	GGCAGAGCATTTTGCTACATCATCTCCGGCCTTCTTCTTCCGCTTA CACAAATGAATCTTAGCTCTCTGCATCGGTTTGTGCAGCTGGGGA TAGTGCTTCGTATCAAAGGACATCGGTGTT	5477
thiolase <i>Cucumis sativus</i> Ser27Term TCG-TAG	AACACCGATGTCCTTTGATACGAAGCACTATCCCCAGCTGCACAA ACCGATGCAGAGAGCTAAGATTCATTTGTGTAAGCGGAAGAAGAA GGCCGGAGATGATGTAGCAAAATGCTCTGCC	5478
ICG-IAG	TGAATCTTAGCTCTCTG	5479
	CAGAGAGCTAAGATTCA	5480
2,4-DB resistance 3-ketoacyl-CoA	TGCTACATCATCTCCGGCCTTCTTCTTCCGCTTACACAAATGAATC TTCGCTCTCTGCATAGGTTTGTGCAGCTGGGGATAGTGCTTCGTA TCAAAGGACATCGGTGTTTGGAGATGATGT	5481
thiolase Cucumis sativus Ser31Term	ACATCATCTCCAAACACCGATGTCCTTTGATACGAAGCACTATCCC CAGCTGCACAAACCTATGCAGAGAGCGAAGATTCATTTGTGTAAG CGGAAGAAGAGGCCGGAGATGATGTAGCA	5482
TCG-TAG	CTCTGCATAGGTTTGTG	5483
	CACAAACC <u>T</u> ATGCAGAG	5484
2,4-DB resistance 3-ketoacyl-CoA	TCATCTCCGGCCTTCTTCTTCCGCTTACACAAATGAATCTTCGCTC TCTGCATCGGTTTGAGCAGCTGGGGATAGTGCTTCGTATCAAAGG ACATCGGTGTTTGGAGATGATGTCGTGATT	5485
thiolase Cucumis sativus Cys33Term	AATCACGACATCATCTCCAAACACCGATGTCCTTTGATACGAAGCA CTATCCCCAGCTGCTCAAACCGATGCAGAGAGCGAAGATTCATTT GTGTAAGCGGAAGAAGAAGCCGGAGATGA	5486
TGT-TGA	TCGGTTTGAGCAGCTGG	548
	CCAGCTGC <u>T</u> CAAACCGA	548
2,4-DB resistance 3-ketoacyl-CoA	GAAGGCAATCAACAGGCAGAGCATTCTGCTACATCATCTCCGGCC TTCATCTTCGGCTTAGAGCCATGAATCTTCGCTCTCTGCATCGGTT TGTGCAGCTGGGGATAGTGCGTCGTATCAA	548
thiolase Cucurbita sp. Tyr22Term TAT-TAG	TTGATACGACGCACTATCCCCAGCTGCACAAACCGATGCAGAGAG CGAAGATTCATGGCTCTAAGCCGAAGATGAAGGCCGGAGATGAT GTAGCAGAATGCTCTGCCTGTTGATTGCCTTC	
IAI-IAG	TCGGCTTAGAGCCATGA	549
	TCATGGCTCTAAGCCGA	549

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	2,4-DB resistance 3-ketoacyl-CoA thiolase	ATCAACAGGCAGAGCATTCTGCTACATCATCTCCGGCCTTCATCTT CGGCTTATAGCCATTAATCTTCGCTCTCTGCATCGGTTTGTGCAG CTGGGGATAGTGCGTCGTATCAAAGAACGT	5493
5	<i>Cucurbita sp.</i> Glu25Term GAA-TAA	ACGTTCTTTGATACGACGCACTATCCCCAGCTGCACAAACCGATG CAGAGAGCGAAGATT <u>A</u> ATGGCTATAAGCCGAAGATGAAGGCCGG AGATGATGTAGCAGAATGCTCTGCCTGTTGAT	5494
		ATAGCCAT <u>T</u> AATCTTCG	5495
		CGAAGATT <u>A</u> ATGGCTAT	5496
	2,4-DB resistance 3-ketoacyl-CoA thiolase	GGCAGAGCATTCTGCTACATCATCTCCGGCCTTCATCTTCGGCTT ATAGCCATGAATCTTAGCTCTCTGCATCGGTTTGTGCAGCTGGGG ATAGTGCGTCGTATCAAAGAACGTCGGTGTT	5497
10	Cucurbita sp. Ser27Term TCG-TAG	AACACCGACGTTCTTTGATACGACGCACTATCCCCAGCTGCACAA ACCGATGCAGAGAGCTAAGATTCATGGCTATAAGCCGAAGATGAA GGCCGGAGATGATGTAGCAGAATGCTCTGCC	5498
•		TGAATCTT <u>A</u> GCTCTCTG	5499
		CAGAGAGC <u>T</u> AAGATTCA	5500
15	2,4-DB resistance 3-ketoacyl-CoA thiolase	TGCTACATCATCTCCGGCCTTCATCTTCGGCTTATAGCCATGAATC TTCGCTCTCTGCATAGGTTTGTGCAGCTGGGGATAGTGCGTCGTA TCAAAGAACGTCGGTGTTTGGAGATGATGT	5501
	Cucurbita sp. Ser31Term TCG-TAG	ACATCATCTCCAAACACCGACGTTCTTTGATACGACGCACTATCCC CAGCTGCACAAACCTATGCAGAGAGCGAAGATTCATGGCTATAAG CCGAAGATGAAGGCCGGAGATGATGTAGCA	5502
		CTCTGCAT <u>A</u> GGTTTGTG	5503
		CACAAACC <u>T</u> ATGCAGAG	5504
20	2,4-DB resistance 3-ketoacyl-CoA thiolase	TCATCTCCGGCCTTCATCTTCGGCTTATAGCCATGAATCTTCGCTC TCTGCATCGGTTTGAGCAGCTGGGGGATAGTGCGTCGTATCAAAGA ACGTCGGTGTTTGGAGATGATGTCGTGATA	5505
	Cucurbita sp. Cys33Term TGT-TGA	TATCACGACATCATCTCCAAACACCGACGTTCTTTGATACGACGCA CTATCCCCAGCTGCTCAAACCGATGCAGAGAGCGAAGATTCATGG CTATAAGCCGAAGATGAAGGCCGGAGATGA	5506
		TCGGTTTGAGCAGCTGG	5507
		CCAGCTGC <u>T</u> CAAACCGA	5508
25	2,4 DB resistance Pex14 Arabidopsis thaliana	TCATAGTCTCTTTTGCCGCTTGGATTCTTCCAAGGTTAGTGAGCTG CTATGGCAACTCATTAGCAAACGCAACCTCCTTCCGATTTTCCCG CTCTTGCCGATGAAAATTCCCAGATTCCAG	5509
	GIn5Term CAG-TAG	CTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAAAATCGGAAG GAGGTTGCGTTTGCT <u>A</u> ATGAGTTGCCATAGCAGCTCACTAACCTT GGAAGAATCCAAGCGGCAAAAGAGACTATGA	5510

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Alteration	CAACTCAT <u>T</u> AGCAAACG	5511
	CGTTTGCT A ATGAGTTG	5512
2,4 DB resistance Pex14	TAGTCTCTTTTGCCGCTTGGATTCTTCCAAGGTTAGTGAGCTGCTA TGGCAACTCATCAGTAAACGCAACCTCCTTCCGATTTTCCCGCTC	5513
Arabidopsis thaliana Gln6Term CAA-TAA	TTGCCGATGAAAATTCCCAGATTCCAGGTT AACCTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAAAATCGG AAGGAGGTTGCGTTTACTGATGAGTTGCCATAGCAGCTCACTAAC CTTGGAAGAATCCAAGCGGCAAAAGAGACTA	5514
·	CTCATCAGTAAACGCAA	5515
	TTGCGTTTACTGATGAG	5516
2,4 DB resistance Pex14	CTTTTGCCGCTTGGATTCTTCCAAGGTTAGTGAGCTGCTATGGCA ACTCATCAGCAAACGTAACCTCCTTCCGATTTTCCCGCTCTTGCC GATGAAAATTCCCAGATTCCAGGTTCAATTT	5517
Arabidopsis thaliana Gln8Term CAA-TAA	AAATTGAACCTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAA AATCGGAAGGAGGTTACGTTTGCTGATGAGTTGCCATAGCAGCTC ACTAACCTTGGAAGAATCCAAGCGGCAAAAG	5518
	ACTAACCTTGGAAGAATCCAAGCGGAVVVC	5519
	AGGAGGTT <u>A</u> CGTTTGCT	5520
2,4 DB resistance Pex14	GCTGCTATGGCAACTCATCAGCAAACGCAACCTCCTTCCGATTTT CCCGCTCTTGCCGATTAAAATTCCCAGATTCCAGGTTCAATTTACA CCTTCTAATCATTATTTCTTAATTTTTCTT	552′
Arabidopsis thaliana Glu19Term GAA-TAA	AAGAAAAATTAAGAAATAATGATTAGAAGGTGTAAATTGAACCTGG AATCTGGGAATTTTAATCGGCAAGAGCGGGAAAATCGGAAGGAG GTTGCGTTTGCTGATGAGTTGCCATAGCAGC	552
	TTGCCGATTAAAATTCC	552
	GGAATTTT A ATCGGCAA	552
2,4 DB resistance Pex14 Arabidopsis thaliana	GCAACTCATCAGCAAACGCAACCTCCTTCCGATTTTCCCGCTCTT GCCGATGAAAATTCCTAGATTCCAGGTTCAATTTACACCTTCTAAT CATTATTTCTTAATTTTTCTTTGGTGGATT	552
GIn22Term CAG-TAG	AATCCACCAAAGAAAATTAAGAAATAATGATTAGAAGGTGTAAATT GAACCTGGAATCTAGGAATTTTCATCGGCAAGAGCGGGAAAATCG GAAGGAGGTTGCGTTTGCTGATGAGTTGC	552
	AAAATTCC <u>T</u> AGATTCCA	552
	TGGAATCT <u>A</u> GGAATTTT	552

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Example 8

Production of albino mutants for the analysis of photosynthetic processes

Plant productivity is limited by resources available and the ability of plants to harness these resources. The conversion of light to chemical energy, which is then used to synthesize carbohydrates, fatty acids, sugars, amino acids and other compounds, requires a complex system which combines the light harvesting apparatus of pigments and proteins. The value of light energy to the plant can only be realized when it is efficiently converted into chemical energy by photosynthesis and fed into various biochemical processes. Significant effort has therefore been directed at studying photosynthetic processes in plants in order to improve productivity and/or the efficiency of photosynthesis. The analysis of the photosynthetic process is substantially aided by the ability to produce albino plants.

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The attached table discloses exemplary oligonucleotide base sequences which can be used to generate site-specific mutations in genes involved in starch metabolism.

Table 18
Oligonucleotides to produce albino plants

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID
White leaves mmutans Arabidopsis thaliana	TTCTTTCCTGTGAAATTATCTGCTCAAATCTTTGGTTCCTGACGGAG ATGGCGGCGATTTGAGGCATCTCCTCTGGTACGTTGACGATTTCA CGGCCTTTGGTTACTCTTCGACGCTCTAG	5529
Ser5Term FCA-TGA	CTAGAGCGTCGAAGAGTAACCAAAGGCCGTGAAATCGTCAACGTA CCAGAGGAGATGCCTCAAATCGCCGCCATCTCCGTCAGGAACCAA AGATTTGAGCAGATAATTTCACAGGAAAGAA	5530
	GGCGATTT <u>G</u> AGGCATCT	5531
	AGATGCCT <u>C</u> AAATCGCC	5532
White leaves Immutans Arabidopsis thaliana	GCTCAAATCTTTGGTTCCTGACGGAGATGGCGGCGATTTCAGGCA TCTCCTCTGGTACGT <u>A</u> GACGATTTCACGGCCTTTGGTTACTCTTCG ACGCTCTAGAGCCGCCGTTTCGTACAGCTC	5533
Leu12Term TTG-TAG	GAGCTGTACGAAACGGCGGCTCTAGAGCGTCGAAGAGTAACCAAA GGCCGTGAAATCGTCTACGTACCAGAGGAGATGCCTGAAATCGCC GCCATCTCCGTCAGGAACCAAAGATTTGAGC	5534
	TGGTACGTAGACGATTT	5535
	AAATCGTC <u>T</u> ACGTACCA	5536
White leaves Immutans Arabidopsis thaliana	TTTGGTTCCTGACGGAGATGGCGGCGATTTCAGGCATCTCCTCTG GTACGTTGACGATTTGACGGCCTTTGGTTACTCTTCGACGCTCTAG AGCCGCCGTTTCGTACAGCTCCTCTCACCG	5537
Ser15Term TCA-TGA	CGGTGAGAGGAGCTGTACGAAACGGCGGCTCTAGAGCGTCGAAG AGTAACCAAAGGCCGT <u>C</u> AAATCGTCAACGTACCAGAGGAGATGCC TGAAATCGCCGCCATCTCCGTCAGGAACCAAA	5538
	GACGATTT <u>G</u> ACGGCCTT	553
·	AAGGCCGT <u>C</u> AAATCGTC	554
White leaves Immutans Arabidopsis thaliana	GCGGCGATTTCAGGCATCTCCTCTGGTACGTTGACGATTTCACGG CCTTTGGTTACTCTTTGACGCTCTAGAGCCGCCGTTTCGTACAGCT CCTCTCACCGATTGCTTCATCATCTTCCTC	554
Arg22Term CGA-TGA	GAGGAAGATGATGAAGCAATCGGTGAGAGGAGCTGTACGAAACG GCGGCTCTAGAGCGTCAAAGAGTAACCAAAGGCCGTGAAATCGTC AACGTACCAGAGGAGATGCCTGAAATCGCCGC	554
	TTACTCTT <u>T</u> GACGCTCT	554
	AGAGCGTC A AAGAGTAA	554

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White leaves mmutans	TCAGGCATCTCCTCTGGTACGTTGACGATTTCACGGCCTTTGGTTA CTCTTCGACGCTCTTGAGCCGCCGTTTCGTACAGCTCCTCTCACC	554
Arabidopsis thaliana	GATTGCTTCATCATCTTCCTCTCTCTCTC	
Arg25Term	GAGAAGAGAGAGAAGATGATGAAGCAATCGGTGAGAGGAGCTG	554
AGA-TGA	TACGAAACGCCGCTCAAGAGCGTCGAAGAGTAACCAAAGGCCG	
NOA-TOA	TGAAATCGTCAACGTACCAGAGGAGATGCCTGA	
	GACGCTCTTGAGCCGCC	554
	GGCGGCTC <u>A</u> AGAGCGTC	554
White leaves	GATTCTTGTGGGAAGGAAGAAGGATCAAGAATGGCGATTTCGATTT	55
Immutans	CTGCTATGAGTTTT <u>T</u> GAACCTCAGTTTCTTCATATTCTTGTTTTAGA	
Lycopersicon	GCTAGGAGTTTTGAGAAGTCATCAGTTT	
esculentum	AAACTGATGACTTCTCAAAACTCCTAGCTCTAAAACAAGAATATGAA	55
Gly11Term	GAAACTGAGGTTC <u>A</u> AAAACTCATAGCAGAAATCGAAATCGCCATTC	
GGA-TGA	TTGATCCTTCCTTCCCACAAGAATC	
	TGAGTTTT <u>T</u> GAACCTCA	55
	TGAGGTTC <u>A</u> AAAACTCA	55
White leaves	GTGGGAAGGAAGAAGGATCAAGAATGGCGATTTCGATTTCTGCTA	55
Immutans	TGAGTTTTGGAACCT <u>G</u> AGTTTCTTCATATTCTTGTTTTAGAGCTAGG	
Lycopersicon	AGTTTTGAGAAGTCATCAGTTTTATGCAA	
esculentum	TTGCATAAAACTGATGACTTCTCAAAACTCCTAGCTCTAAAACAAGA	55
Ser13Term	ATATGAAGAAACT <u>C</u> AGGTTCCAAAACTCATAGCAGAAATCGAAATC	
TCA-TGA	GCCATTCTTGATCCTTCCTTCCCAC	
	TGGAACCT <u>G</u> AGTTTCTT	55
	AAGAAACT <u>C</u> AGGTTCCA	55
White leaves	AAGAAGGATCAAGAATGGCGATTTCGATTTCTGCTATGAGTTTTGG	55
Immutans	AACCTCAGTTTCTT <u>G</u> ATATTCTTGTTTTAGAGCTAGGAGTTTTGAGA	
Lycopersicon	AGTCATCAGTTTTATGCAATTCCCAGAA	
esculentum	TTCTGGGAATTGCATAAAACTGATGACTTCTCAAAACTCCTAGCTC	55
Ser16Term	TAAAACAAGAATAT <u>C</u> AAGAAACTGAGGTTCCAAAACTCATAGCAGA	
TCA-TGA	AATCGAAATCGCCATTCTTGATCCTTCTT	
	AGTTTCTT <u>G</u> ATATTCTT	55
	AAGAATAT <u>C</u> AAGAAACT	55
White leaves	AGGATCAAGAATGGCGATTTCGATTTCTGCTATGAGTTTTGGAACC	55
Immutans	TCAGTTTCTTCATAGTCTTGTTTTAGAGCTAGGAGTTTTGAGAAGTC	
Lycopersicon	ATCAGTTTTATGCAATTCCCAGAACCCA	
esculentum	TGGGTTCTGGGAATTGCATAAAACTGATGACTTCTCAAAACTCCTA	55
Tyr17Term	GCTCTAAAACAAGA <u>C</u> TATGAAGAAACTGAGGTTCCAAAACTCATAG	
TAT-TAG	CAGAAATCGAAATCGCCATTCTTGATCCT	<u> </u>
	TCTTCATA <u>G</u> TCTTGTTT	55
	AAACAAGA C TATGAAGA	55

	White leaves	AAGAATGGCGATTTCGATTTCTGCTATGAGTTTTGGAACCTCAGTT TCTTCATATTCTTGATTTAGAGCTAGGAGTTTTGAGAAGTCATCAGT	5565
	Immutans Lycopersicon esculentum	TTTATGCAATTCCCAGAACCCATGTCGG CCGACATGGGTTCTGGGAATTGCATAAAACTGATGACTTCTCAAAA CTCCTAGCTCTAAATCAAGAATATGAAGAAACTGAGGTTCCAAAAC	5566
	Cys19Term TGT-TGA	TCATAGCAGAAATCGAAATCGCCATICTT	5567
		TATTCTTGATTTAGAGC GCTCTAAATCAAGAATA	5568
	White leaves	CGCGTCCGATAAAAAAATCAAGAATGGCGATTTCCATATCTGCTAT GAGTTTTCGAACTT G AGTTTCTTCTTCATATTCAGCATTTTTGTGCA	5569
	Capsicum annuum Ser13Term TCA-TGA	ATTCCAAGAACCCATTTTGTTTGAATTC GAATTCAAACAAAATGGGTTCTTGGAATTGCACAAAAATGCTGAAT ATGAAGAAGAAACTCAAGTTCGAAAACTCATAGCAGATATGGAAAT	5570
		CGCCATTCTTGATTTTTTTATCGGACGCG TCGAACTTGAGTTTCTT	5571
		AAGAAACT C AAGTTCGA	5572
	White leaves Immutans	AAAAATCAAGAATGGCGATTTCCATATCTGCTATGAGTTTTCGAACT TCAGTTTCTTCTTGATATTCAGCATTTTTGTGCAATTCCAAGAACCC ATTTTGTTTGAATTCTCTATTTTCACT	5573
;	Capsicum annuum Ser17Term TCA-TGA	AGTGAAAATAGAGAATTCAAACAAAATGGGTTCTTGGAATTGCACA AAAATGCTGAATATCAAGAAGAAACTGAAGTTCGAAAACTCATAGC	5574
		AGATATGGAAATCGCCATTCTTGATTTTT TTCTTCTTGATATTCAG	5575
		CTGAATAT <u>C</u> AAGAAGAA	5576
	White leaves Immutans	CAAGAATGGCGATTTCCATATCTGCTATGAGTTTTCGAACTTCAGT TTCTTCTTCATATT G AGCATTTTTGTGCAATTCCAAGAACCCATTTT	5577
0	Capsicum annuum Ser19Term TCA-TGA	GTTTGAATTCTCTATTTTCACTTAGGAA TTCCTAAGTGAAAATAGAGAAATTCAAACAAAATGGGTTCTTGGAATT GCACAAAAATGCTCAATATGAAGAAGAAACTGAAGTTCGAAAACTC	5578
		ATAGCAGATATGGAAATCGCCATTCTTG TTCATATTGAGCATTTT	5579
		AAAATGCT C AATATGAA	5580
	White leaves Immutans	CGATTTCCATATCTGCTATGAGTTTTCGAACTTCAGTTTCTTCA TATTCAGCATTTTAGTGCAATTCCAAGAACCCATTTTGTTTG	5581
25	Capsicum annuum Leu21Term TTG-TAG	TCTATTTCACTTAGGAATTCTCATAG CTATGAGAATTCCTAAGTGAAAATAGAGAATTCAAACAAA	5582
		CGAAAACTCATAGCAGATATGGAAATCG AGCATTTTAGTGCAATT	558
		AATTGCAC <u>T</u> AAAATGCT	558

	White leaves Immutans	TTCCATATCTGCTATGAGTTTTCGAACTTCAGTTTCTTCATATT CAGCATTTTTGTGAAATTCCAAGAACCCATTTTGTTTGAATTCTCTA	5585
5	Capsicum annuum Cys22Term TGC-TGA	TTTTCACTTAGGAATTCTCATAGAACT AGTTCTATGAGAATTCCTAAGTGAAAATAGAGAATTCAAACAAA	5586
		TTTTGTG <u>A</u> AATTCCAA	5587
		TTGGAATT <u>T</u> CACAAAAA	5588
	White leaves Immutans Oryza sativa	TTCGGCACGAGGAGAAGGAGCAGACCGAGGTGGCCGTCGAGG AGTCCTTCCCCTTCAGGTAGACGGCTCCTCCTGACGAGCCACTGG TCACCGCCGAGGAGAGCTGGGTGGTTAAGCTCG	5589
10	Glu22Term GAG-TAG	CGAGCTTAACCACCCAGCTCTCCTCGGCGGTGACCAGTGGCTCG TCAGGAGGAGCCGTCTACCTGAAGGGGAAGGACTCCTCGACGGC CACCTCGGTCTGCTCCTTCTCCCTCGTGCCGAA	5590
		CCTTCAGG <u>T</u> AGACGGCT	5591
		AGCCGTCT <u>A</u> CCTGAAGG	5592
	White leaves Immutans Oryza sativa	GAGCAGACCGAGGTGGCCGTCGAGGAGTCCTTCCCCTTCAGGGA GACGGCTCCTCGACTAGCCACTGGTCACCGCCGAGGAGAGCT GGGTGGTTAAGCTCGAGCAGTCCGTGAACATTT	5593
15	Glu28Term CAG-TAG	AAATGTTCACGGACTGCTCGAGCTTAACCACCCAGCTCTCCTCGG CGGTGACCAGTGGCTAGTCAGGAGGAGCCGTCTCCCTGAAGGGG AAGGACTCCTCGACGGCCACCTCGGTCTGCTC	5594
		CTCCTGAC <u>T</u> AGCCACTG	5595
		CAGTGGCT <u>A</u> GTCAGGAG	5596
	White leaves Immutans Oryza sativa	GTCGAGGAGTCCTTCCCCTTCAGGGAGACGGCTCCTCCTGACGA GCCACTGGTCACCGCCTAGGAGAGCTGGGTGGTTAAGCTCGAGC AGTCCGTGAACATTTTCCTCACGGAGTCAGTCA	5597
20	Glu34Term GAG-TAG	TGACTGACTCCGTGAGGAAAATGTTCACGGACTGCTCGAGCTTAA CCACCCAGCTCTCCTAGGCGGTGACCAGTGGCTCGTCAGGAGGA GCCGTCTCCCTGAAGGGGAAGGACTCCTCGAC	5598
		TCACCGCC <u>T</u> AGGAGAGC	5599
		GCTCTCCT <u>A</u> GGCGGTGA	5600
	White leaves Immutans Oryza sativa	GAGGAGTCCTTCCCCTTCAGGGAGACGGCTCCTCCTGACGAGCC ACTGGTCACCGCCGAGTAGAGCTGGGTGATTAAGCTCGAGCAGT CCGTGAACATTTTCCTCACGGAGTCAGTCATCA	5601
25	Glu35Term GAG-TAG	TGATGACTGACTCCGTGAGGAAAATGTTCACGGACTGCTCGAGCT TAACCACCCAGCTCTACTCGGCGGTGACCAGTGGCTCGTCAGGA GGAGCCGTCTCCCTGAAGGGGAAGGACTCCTC	5602
		CCGCCGAGTAGAGCTGG	5603
		CCAGCTCT <u>A</u> CTCGGCGG	5604

	White leaves Immutans	CGCCGAGGAGACTGAGCTCGAGCAGTCCGTGAACA	5605
5	Oryza sativa Trp37Term TGG-TGA	TTTTCCTCACGGAGTCAGTCATCACGATACTT AAGTATCGTGATGACTGACTCCGTGAGGAAAATGTTCACGGACTG CTCGAGCTTAACCACTCAGCTCTCCTCGGCGGTGACCAGTGGCTC	5606
		GTCAGGAGGAGCCGTCTCCCTGAAGGGGAAG GAGAGCTGAGTGATAA	5607
		TTAACCAC <u>T</u> CAGCTCTC	5608
	White leaves Immutans	TCCGGAGGAGGAGGGGGGTTCGACGAGGAGCTCACCCTCGCCG GCGAGGACGGCGACTGAGTCGTCAGATTCGAGCAGTCCTTCAAC GTATTCCTCACGGATACTGTCATCTTTATACTC	5609
10	Triticum aestivum Trp22Term TGG-TGA	GAGTATAAAGATGACAGTATCCGTGAGGAATACGTTGAAGGACTG CTCGAATCTGACGACTCAGTCGCCGTCCTCGCCGGCGAGGGTGA GCTCCTCGTCGAATCCCCCTTCCTCCGGA	5610
		GGCGACTG <u>A</u> GTCGTCAG	5611
		CTGACGAC <u>T</u> CAGTCGCC	5612
	White leaves Immutans	GAGGAAGGGGATTCGACGAGGAGCTCACCCTCGCCGGCGAGG ACGGCGACTGGGTCGTCTGATTCGAGCAGTCCTTCAACGTATTCC TCACGGATACTGTCATCTTTATACTCGATATTC	5613
15	Triticum aestivum Arg25Term AGA-TGA	GAATACTGTCATOTTTATAGESCAGAATACGTTGAA GAATATCGAGTATAAAGATGACAGTATCCGTGAGGAATACGTTGAA GGACTGCTCGAATCAGACCAGTCGCCGTCCTCGCCGGCGA GGGTGAGCTCCTCGTCGAATCCCCCTTCCTC	5614
		GGGTGAGCTCCTCGTCGAATCCCCCTTGGTG GGGTCGTCTCGAG	5615
		CTCGAATCAGACGACCC	5616
	White leaves Immutans	GGGGGATTCGACGAGGAGCTCACCCTCGCCGGCGAGGACGGCG ACTGGGTCGTCAGATTCTAGCAGTCCTTCAACGTATTCCTCACGGA TACTGTCATCTTTATACTCGATATTCTGTATC	5617
20	Triticum aestivum Glu27Term GAG-TAG	GATACAGAATATCGAGTATAAAGATGACAGTATCCGTGAGGAATAC GTTGAAGGACTGCTAGAATCTGACGACCCAGTCGCCGTCCTCGCC GGCGAGGGTGAGCTCCTCGTCGAATCCCCC	5618
•		TCAGATTCTAGCAGTCC	5619
		GGACTGCTAGAATCTGA	5620
	White leaves Immutans	GGATTCGACGAGGAGCTCACCCTCGCCGGCGAGGACGGCGACTG GGTCGTCAGATTCGAGTAGTCCTTCAACGTATTCCTCACGGATACT	
25	Triticum aestivum Gln28Term CAG-TAG	CACGATACAGAATATCGAGTATAAAGATGACAGTATCCGTGAGGAA TACGTTGAAGGACTACTCGAATCTGACGACCCAGTCGCCGTCCTC GCCGGCGAGGGTGAGCTCCTCGTCGAATCC	
		GATTCGAGTAGTCCTTC	5623
		GAAGGACTACTCGAATC	5624

White leaves	CGAGCAGTCCTTCAACGTATTCCTCACGGATACTGTCATCTTTATA	5625
Immutans	CTCGATATTCTGTA <u>G</u> CGTGACCGCGACTACGCAAGGTTCTTCGTG	
Triticum aestivum	CTCGAGACCATCGCCAGGGTGCCCTATTTC	
Tyr46Term	GAAATAGGGCACCCTGGCGATGGTCTCGAGCACGAAGAACCTTG	5626
TAT-TAG	CGTAGTCGCGGTCACG <u>C</u> TACAGAATATCGAGTATAAAGATGACAG	
	TATCCGTGAGGAATACGTTGAAGGACTGCTCG	
	ATTCTGTA <u>G</u> CGTGACCG	5627
	CGGTCACG <u>C</u> TACAGAAT	5628

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Example 9

Altering amino acid content of plants

Another aim of biotechnology is to generate plants, especially crop plants, with added value traits. An example of such a trait is improved nutritional quality in food crops. For example, lysine, tryptophan and threonine, which are essential amino acids in the diet of humans and many animals, are limiting nutrients in most cereal crops. Consequently, grain-based diets, such as those based on corn, barley, wheat, rice, maize, millet, sorghum, and the like, must be supplemented with more expensive synthetic amino acids or amino-acid-containing oilseed protein meals. Increasing the lysine content of these grains or of any of the feed component crops would result in significant added value.

Naturally occurring mutants of plants that have different levels of particular essential amino acids have been identified. However, these mutants are generally not the result of increased free amino acid, but are instead the result of shifts in the overall protein profile of the grain. For example, in maize, reduced levels of lysine-deficient endosperm proteins (prolamines) are complemented by elevated levels of more lysine-rich proteins (albumins, globulins and glutelins). While nutritionally superior, these mutants are associated with reduced yields and poor grain quality, limiting their agronomic usefulness.

An alternative approach is to generate plants with mutations that render key amino acid biosynthetic enzymes insensitive to feedback inhibition. Many such mutations are known and mutation results in increased free amino acid. The increased production can optionally be coupled to increased expression of an abundant storage protein comprising the chosen amino acid. Alternatively, a normally abundant protein can be engineered to contain more of the target amino acid.

The attached table discloses exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that remove feedback inhibition in plant amino acid biosynthetic enzymes.

Table 19 **Genome-Altering Oligos Conferring Amino Acid Overproduction**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Met Overproduction CGS Arabidopsis thaliana	TATCCTCCAGGATCTTAAGATTTCCTCCTAATTTCGTCCGTC	5629
Arg77His CGT-CAT	GGGTTGTTGGACCACTTAGCCGCCACGATCTGTGCAACACCGAT GTTGCTACAGTTTCTATGGGCTTTAATGCTCAGCTGACGGACG	5630
	TAAAGCCC <u>A</u> TAGAAACT	5631
	AGTTTCTA <u>T</u> GGGCTTTA	5632
Met Overproduction CGS Arabidopsis thaliana	TCTTAAGATTTCCTCCTAATTTCGTCCGTCAGCTGAGCATTAAAGC CCGTAGAAACTGTAACAACATCGGTGTTGCACAGATCGTGGCGG CTAAGTGGTCCAACAACCCATCCTCCGCGTT	5633
Ser81Asn AGC-AAC	AACGCGGAGGATGGGTTGTTGGACCACTTAGCCGCCACGATCTG TGCAACACCGATGTTGTTACAGTTTCTACGGGCTTTAATGCTCAGC TGACGGACGAAATTAGGAGGAAATCTTAAGA	5634
	AAACTGTA <u>A</u> CAACATCG	5635
	CGATGTTG <u>T</u> TACAGTTT	5636
Met Overproduction CGS Arabidopsis thaliana	TTTCCTCCTAATTTCGTCCGTCAGCTGAGCATTAAAGCCCGTAGAA ACTGTAGCAACATCAGTGTTGCACAGATCGTGGCGGCTAAGTGGT CCAACAACCCATCCTCCGCGTTACCTTCGG	5637
Gly84Ser GGT-AGT	CCGAAGGTAACGCGGAGGATGGGTTGTTGGACCACTTAGCCGCC ACGATCTGTGCAACACTGATGTTGCTACAGTTTCTACGGGCTTTAA TGCTCAGCTGACGGACGAAATTAGGAGGAAA	5638
	GCAACATC <u>A</u> GTGTTGCA	5639
	TGCAACAC <u>T</u> GATGTTGC	5640
Met Overproduction CGS Arabidopsis thaliana	TTCCTCCTAATTTCGTCCGTCAGCTGAGCATTAAAGCCCGTAGAAA CTGTAGCAACATCGATGTTGCACAGATCGTGGCGGCTAAGTGGTC CAACAACCCATCCTCCGCGTTACCTTCGGC	5641
Gly84Asp GGT-GAT	GCCGAAGGTAACGCGGAGGATGGGTTGTTGGACCACTTAGCCGC CACGATCTGTGCAACATCGATGTTGCTACAGTTTCTACGGGCTTTA ATGCTCAGCTGACGGACGAAATTAGGAGGAA	5642
	CAACATCG <u>A</u> TGTTGCAC	5643
	GTGCAACA <u>T</u> CGATGTTG	5644

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Met Overproduction CGS	TATCGTCACTCATCCTCCGCTTCCCTCCCAACTTCGTCCGCCAGC TCAGCACCAAGGCCCACCGCAACTGCAGCAACATCGCGCGTCGCG CAGATCGTCGCGGCTTCGTGGTCCAACAAGA	5645
Fragraria vesca Arg73His CGC-CAC	TCTTTGTTGGACCACGAAGCCGCGACGATCTGCGCGACGCCGAT GTTGCTGCAGTTGCGGTGGGCCTTGGTGCTGAGCTGGCGGACGA AGTTGGGAGGGAAGCGGAGGATGAGTGACGATA	5646
	CAAGGCCCACCGCAACT	5647
	AGTTGCGGTGGGCCTTG	5648
Met Overproduction CGS	TCCTCCGCTTCCCCAACTTCGTCCGCCAGCTCAGCACCAAG GCCCGCCGCAACTGCAACAACATCGGCGTCGCGCAGATCGTCGC GGCTTCGTGGTCCAACAAGACTCCGACCTTTC	5649
Fragraria vesca Ser77Asn AGC-AAC	GAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGCGACGATCTG CGCGACGCCGATGTTGTTGCAGTTGCGGCGGGCCTTGGTGCTGA GCTGGCGGACGAAGTTGGGAGGGAAGCGGAGGA	5650
	CAACTGCAACATCG	5651
	CGATGITGTGCAGTTG .	5652
Met Overproduction CGS	TTCCCTCCAACTTCGTCCGCCAGCTCAGCACCAAGGCCCGCCG CAACTGCAGCAACATCAGCGTCGCGCAGATCGTCGCGGCTTCGT GGTCCAACAAGACTCCGACCTTTCGGCGGTGC	5653
Fragraria vesca Gly80Ser GGC-AGC	GCACCGCGAAAGGTCGGAGCTTTGCGGCGCAAGCCGCG GCACCGCGAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGCG ACGATCTGCGCGACGACGATGTTGCTGCAGTTGCGGCGGGCCTT GGTGCTGAGCTGGCGGACGAAGTTGGGAGGGAA	5654
	GCAACATCAGCGTCGCG	5655
	CGCGACGC <u>T</u> GATGTTGC	5656
Met Overproduction CGS	TCCCTCCAACTTCGTCCGCCAGCTCAGCACCAAGGCCCGCCGCAAACACACAC	565
Fragraria vesca Gly80Asp GGC-GAC	GGCACCGCGAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGC GACGATCTGCGCGACG <u>T</u> CGATGTTGCTGCAGTTGCGGCGGGCCT TGGTGCTGAGCTGGCGGACGAAGTTGGGAGGGA	565
	CAACATCG <u>A</u> CGTCGCGC	565
	GCGCGACGTCGATGTTG	566
Met Overproduction CGS	TCTCCTCCTCATCCTCCGCTTCCCTCCCAACTTCCAGCGCCAGC TAAGCACCAAGGCGAGCCGCAACTGCAGCAACATCGCGCGTCGCG CAAATCGTCGCCGCTTCGTGGTCGAACAACAG	566
Glycine max Arg68His CGC-CAC	CTGTTGTTCGCCGCTTCGTGGTGCACACTTGCGCGACGCCGAT GTTGCTGCAGTTGCGGCTCGCCTTGGTGCTTAGCTGGCGCTGGA AGTTGGGAGGGAAGCGGAGGATGAGGGAGGAGA	566
	CCAAGGCGAGCCGCAAC	566

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GTTGCGGC <u>T</u> CGCCTTGG	5664
Met Overproduction CGS Glycine max	TCCTCCGCTTCCCCAACTTCCAGCGCCAGCTAAGCACCAAG GCGCGCCGCAACTGCAACAACATCGGCGTCGCGCAAATCGTCGC CGCTTCGTGGTCGAACAACAGCGACAACTCTCC	5665
Ser72Asn AGC-AAC	GGAGAGTTGTCGCTGTTGTTCGACCACGAAGCGGCGACGATTTG CGCGACGCCGATGTTGTTGCAGTTGCGGCGCGCCCTTGGTGCTTA GCTGGCGCTGGAAGTTGGGAGGGAAGCGGAGGA	5666
	CAACTGCA <u>A</u> CAACATCG	5667
	CGATGTTGTTGCAGTTG	5668
Met Overproduction CGS Glycine max	TTCCCTCCAACTTCCAGCGCCAGCTAAGCACCAAGGCGCGCCG CAACTGCAGCAACATCAGCGTCGCGCAAATCGTCGCCGCTTCGT GGTCGAACAACAGCGACAACTCTCCGGCCGCCG	5669
Gly75Ser GGC-AGC	CGGCGGCCGAGAGTTGTCGCTGTTGTTCGACCACGAAGCGGC GACGATTTGCGCGACGC <u>T</u> GATGTTGCTGCAGTTGCGGCGCGCCT TGGTGCTTAGCTGGCGCTGGAAGTTGGGAGGGAA	5670
	GCAACATC <u>A</u> GCGTCGCG	5671
	CGCGACGC <u>T</u> GATGTTGC	5672
Met Overproduction ĆGS Glycine max	TCCCTCCCAACTTCCAGCGCCAGCTAAGCACCAAGGCGCGCCGC AACTGCAGCAACATCGACGTCGCGCAAATCGTCGCCGCTTCGTG GTCGAACAACAGCGACAACTCTCCGGCCGCCGG	5673
Gly75Asp GGC-GAC	CCGGCGGCCGAGAGTTGTCGCTGTTGTTCGACCACGAAGCGGC GACGATTTGCGCGACG <u>T</u> CGATGTTGCTGCAGTTGCGGCGCGCCCT TGGTGCTTAGCTGGCGCTGGAAGTTGGGAGGGA	5674
	CAACATCGACGTCGCGC	5675
	GCGCGACG <u>T</u> CGATGTTG	5676
Met Overproduction CGS Solanum tuberosum	TGTCTTCTCGATTTTCAGGTTTCCTCCTAATTTCGTGAGGCAGCT ,AAGCATTAAGGCT <u>CAC</u> AGGAATTGCAGCAATATTGGCGTGGCTCA AGTTGTGGCGGCTTCCTGGTCTAACAACCA	5677
Arg70His AGG-CAC	TGGTTGTTAGACCAGGAAGCCGCCACACTTGAGCCACGCCAATA TTGCTGCAATTCCT GTG AGCCTTAATGCTTAGCTGCCTCACGAAAT TAGGAGGAAACCTGAAAATCAGAGAAGACA	5678
	TAAGGCT <u>CAC</u> AGGAATT	5679
	AATTCCT GTG AGCCTTA	5680
Met Overproduction CGS Solanum tuberosum	TTTTCAGGTTTCCTCCTAATTTCGTGAGGCAGCTAAGCATTAAGGC TAGGAGGAATTGCAACAATATTGGCGTGGCTCAAGTTGTGGCGG CTTCCTGGTCTAACAACCAAGCCGGTCCTGA	5681
Ser74Asn AGC-AAC	TCAGGACCGGCTTGGTTGTTAGACCAGGAAGCCGCCACAACTTG AGCCACGCCAATATTGTTGCAATTCCTCCTAGCCTTAATGCTTAGC TGCCTCACGAAATTAGGAGGAAACCTGAAAA	5682

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GAATTGCA <u>A</u> CAATATTG	5683
	CAATATTG <u>T</u> TGCAATTC	5684
Met Overproduction CGS	TTTCCTCCTAATTTCGTGAGGCAGCTAAGCATTAAGGCTAGGAGG AATTGCAGCAATATTAGCGTGGCTCAAGTTGTGGCGGCTTCCTGG TCTAACAACCAAGCCGGTCCTGAATTCACTC	5685
Solanum tuberosum Gly77Ser GGC-AGC	GAGTGAATTCAGGACCGGCTTGATTGATCAGACCAGGAAGCCGCC ACAACTTGAGCCACGCTAATATTGCTGCAATTCCTCCTAGCCTTAA TGCTTAGCTGCCTCACGAAATTAGGAGGAAA	5686
	GCAATATT <u>A</u> GCGTGGCT	5687
	AGCCACGCTAATATTGC	5688
Met Overproduction CGS	TTCCTCCTAATTTCGTGAGGCAGCTAAGCATTAAGGCTAGGAGGA ATTGCAGCAATATTGACGTGGCTCAAGTTGTGGCGGCTTCCTGGT CTAACAACCAAGCCGGTCCTGAATTCACTCC	5689
Solanum tuberosum Gly77Asp GGC-GAC	GGAGTGAATTCAGGACCGGCTTGGTTGTTAGACCAGGAAGCCGC CACAACTTGAGCCACGTCAATATTGCTGCAATTCCTCCTAGCCTTA ATGCTTAGCTGCCTCACGAAATTAGGAGGAA	5690
	CAATATTGACTGGCTC	5691
	GAGCCACGTCAATATTG	5692
Met Overproduction CGS	CTTCCTCTTATCCTTCGCTTTCCTCCCAACTTTGTCCGTCAGCT CAGCACCAAGGCTCGCCACAACTGCAGCAACATTGGTGTCGCAC AGGTCGTCGCTGCCTCCTGGTCCAACAACTC	5693
Mesembryanthemum crystallinum Arg73His	GAGTTGTTGGACCAGGAGGCAGCGACGACCTGTGCGACACCAAT GTTGCTGCAGTTGTGGCGAGCCTTGGTGCTGAGCTGA	5694
CGC-CAC	GGCTCGCCACAACTGCA	5695
	TGCAGTTG <u>T</u> GGCGAGCC	5696
Met Overproduction CGS Mesembryanthemum	TCCTTCGCTTTCCTCCCAACTTTGTCCGTCAGCTCAGCACCAAGG CTCGCCGCAACTGCAACAACATTGGTGTCGCACAGGTCGTCGCT GCCTCCTGGTCCAACAACTCCGATGCCGGCGC	5697
crystallinum Ser77Asn AGC-AAC	GCGCCGCATCGGAGTTGTTGGACCAGGAGGCAGCGACCT GTGCGACACCAATGTTGTTGCAGTTGCGGCGAGCCTTGGTGCTG AGCTGACGGACAAAGTTGGGAGGAAAGCGAAGGA	5698
700-770	CAACTGCAACAACATTG	5699
	CAATGTTGTTGCAGTTG	570

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Met Overproduction CGS Mesembryanthemum	TTTCCTCCCAACTTTGTCCGTCAGCTCAGCACCAAGGCTCGCCGC AACTGCAGCAACATTAGTGTCGCACAGGTCGTCGCTGCCTCCTG GTCCAACAACTCCGATGCCGGCGCCACCTCTT	5701
crystallinum Gly80Ser GGT-AGT	AAGAGGTGGCGCCGGCATCGGAGTTGTTGGACCAGGAGGCAGC GACGACCTGTGCGACAC <u>T</u> AATGTTGCTGCAGTTGCGGCGAGCCT TGGTGCTGACCTGAC	5702
	GCAACATT <u>A</u> GTGTCGCA	5703
	TGCGACAC <u>T</u> AATGTTGC	5704
Met Overproduction CGS Mesembryanthemum	TTCCTCCCAACTTTGTCCGTCAGCTCAGCACCAAGGCTCGCCGCA ACTGCAGCAACATTGATGTCGCACAGGTCGTCGCTGCCTCCTGG TCCAACAACTCCGATGCCGGCGCCACCTCTTG	5705
crystallinum Gly80Asp GGT-GAT	CAAGAGGTGCCCCGGCATCGGAGTTGTTGGACCAGGAGGCAG CGACGACCTGTGCGACA <u>T</u> CAATGTTGCTGCAGTTGCGGCGAGCC TTGGTGCTGAGCTGA	5706
•	CAACATTG <u>A</u> TGTCGCAC	5707
	GTGCGACA <u>T</u> CAATGTTG	5708
Met Overproduction CGS Zea mays	CCTCTGCTACCATCCTCCGCTTTCCGCCAAACTTTGTCCGCCAGC TTAGCACCAAGGCACAACCACCGCGCGCGCGCGCGCGCGC	5709
Arg41His CGC-CAC	GGGCAGTCGGACCACGCGGCGGCGACGATCTGCGCGACGCCGA TGTTGCTGCAGTTGCGGTGTGCCTTGGTGCTAAGCTGGCGGACA AAGTTTGGCGGAAAGCGGAGGATGGTAGCAGAGG	5710
	CAAGGCAC <u>A</u> CCGCAACT	5711
. •	AGTTGCGG <u>T</u> GTGCCTTG	5712
Met Overproduction CGS Zea mays	TCCTCCGCTTTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGG CACGCCGCAACTGCAACATCGGCGTCGCGCAGATCGTCGCC GCCGCGTGGTCCGACTGCCCCGCCGCTCGCCC	5713
Ser45Asn AGC-AAC	GGGCGAGCGGGGGCAGTCGGACCACGCGGCGCGACGATC TGCGCGACGCCGATGTTGTTGCAGTTGCGGCGTGCCTTGGTGCT AAGCTGGCGGACAAAGTTTGGCGGAAAGCGGAGGA	5714
	CAACTGCA <u>A</u> CAACATCG	5715
	CGATGTTG <u>T</u> TGCAGTTG	5716
Met Overproduction CGS Zea mays	TTTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGGCACGCCGC AACTGCAGCAACATCAGCGTCGCGCAGATCGTCGCCGCGCGTG GTCCGACTGCCCCGCCGCCGCCCCCCCCCC	5717
Gly48Ser GGC-AGC	CTAAGTGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGG	5718
	GCAACATCAGCGTCGCG	5719

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ II NO:
Anteration	CGCGACGC <u>T</u> GATGTTGC	5720
Met Overproduction CGS	TTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGGCACGCCGC AACTGCAGCAACATCGACGTCGCGCAGATCGTCGCCGCGCGTG	5721
Zea mays Gly48Asp GGC-GAC	GTCCGACTGCCCCGCCGCCCCCACTTAGG CCTAAGTGGGGGCGAGCGGGGGGGCAGTCGGACCACGCGGCG GCGACGATCTGCGCGACGTCGATGTTGCTGCAGTTGCGGCGTGC CTTGGTGCTAAGCTGGCGGACAAAGTTTGGCGGAA	5722
	CAACATCGACGTCGCGC	5723
	GCGCGACGTCGATGTTG	572
Met Overproduction TS	GTATGAATGATCTGTGGGTGAAACACTGTGGGATTAGTCATACAG GAAGTTTCAAGGATCGTGGAATGACTGTTTTGGTTAGTCAAGTTAA TCGTCTGAGAAAGATGAAACGACCTGTGGT	572
Arabidopsis thaliana Leu205Arg CTT-CGT	ACCACAGGTCGTTTCATCTTTCTCAGACGATTAACTTGACTAACCA AAACAGTCATTCCACGATCCTTGAAACTTCCTGTATGACTAATCCC ACAGTGTTTCACCCACAGATCATTCATAC	572
	CAAGGATCGTGGAATGA	572
	TCATTCCACGATCCTTG	572
Met Overproduction TS	GCATGACTGATTTGTGGGTCAAACACTGTGGGATTAGCCATACTG GTAGTTTTAAGGATCGTGGGATGACTGTTTTGGTGAGTCAAGTTAA TCGCTTGCGGAAAATGCATAAACCGGTTGT	572
Solanum tuberosum Leu198Arg CTT-CGT	ACAACCGGTTTATGCATTTTCCGCAAGCGATTAACTTGACTCACCA AAACAGTCATCCCACGATCCTTAAAACTACCAGTATGGCTAATCCC ACAGTGTTTGACCCACAAATCAGTCATGC	573
	TAAGGATCGTGGGATGA	57
	TCATCCCACGATCCTTA	57
Lys Overproduction DHPS	TCATTGGGCACACAGTGAACTGCTTTGGCTCTAGAATCAAAGTGA TAGGCAACACAGGAALTGCTGTTGGCATGCATGC ACACACAGGGATTTGCTGTTGGCATGCATGC	57
Zea mays Ser157Asn AGC-AAC	GCATGCATGCCAACAGCAAATCCCTGTTCTGTTGCGTGGACGGCT TCTCTGGTTGAGTTGTTTCCTGTGTTGCCTATCACTTTGATTCTAG AGCCAAAGCAGTTCACTGTGTGCCCAATGA	57
	CACAGGAAACAACTCAA	57
	TTGAGTTGTTTCCTGTG	57
Lys Overproduction DHPS		
Zea mays Ala166Val GCA-GAA	CCGTAGTAAGGATTGATGTGGAGAGCCGCATGCATGCCAACAGC AAATCCCTGTTCTGTT	5

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CGTCCACG <u>A</u> AACAGAAC	5739
	GTTCTGTT <u>T</u> CGTGGACG	5740
Lys Overproduction DHPS Zea mays	GGCTCTAGAATCAAAGTGATAGGCAACACAGGAAGCAACTCAACC AGAGAAGCCGTCCACACAACAGAACAG	5741
Ala166Thr GCA-ACA	CGTAGTAAGGATTGATGTGGAGAGCCGCATGCATGCCAACAGCAA ATCCCTGTTCTGTT	5742
İ	CCGTCCAC <u>A</u> CAACAGAA	5743
	TTCTGTTG <u>T</u> GTGGACGG	5744
Lys Overproduction DHPS Oryza sativa	TTATTGGGCATACAGTTAACTGCTTTGGCACTAAAATTAAAGTGGT CGGCAACACAGGAAATAACTCAACAAGGGAGGCTATTCACGCAAC TGAGCAGGGATTCGCTGTAGGTATGCACGC	5745
Ser124Asn AGT-AAT	GCGTGCATACCTACAGCGAATCCCTGCTCAGTTGCGTGAATAGCC TCCCTTGTTGAGTTA <u>T</u> TTCCTGTGTTGCCGACCACTTTAATTTTAGT GCCAAAGCAGTTAACTGTATGCCCAATAA	5746
	CACAGGAA <u>A</u> TAACTCAA	5747
	TTGAGTTATTTCCTGTG	5748
Lys Overproduction DHPS Oryza sativa	GCACTAAAATTAAAGTGGTCGGCAACACAGGAAGTAACTCAACAA GGGAGGCTATTCACG <u>T</u> AACTGAGCAGGGATTCGCTGTAGGTATG CACGCGGCTCTCCACATCAATCCTTACTACGG	5749
Ala133Val GCA-GTA	CCGTAGTAAGGATTGATGTGGAGAGCCGCGTGCATACCTACAGC GAATCCCTGCTCAGTTACGTGAATAGCCTCCCTTGTTGAGTTACTT CCTGTGTTGCCGACCACTTTAATTTTAGTGC	5750
	TATTCACG <u>T</u> AACTGAGC	5751
	GCTCAGTT <u>A</u> CGTGAATA	5752
Lys Overproduction DHPS Oryza sativa	GGCACTAAAATTAAAGTGGTCGGCAACACAGGAAGTAACTCAACA AGGGAGGCTATTCACACAACTGAGCAGGGATTCGCTGTAGGTAT GCACGCGGCTCTCCACATCAATCCTTACTACG	5753
Ala133Thr GCA-ACA	CGTAGTAAGGATTGATGTGGAGAGCCGCGTGCATACCTACAGCG AATCCCTGCTCAGTTGTGTGAATAGCCTCCCTTGTTGAGTTACTTC CTGTGTTGCCGACCACTTTAATTTTAGTGCC	5754
	CTATTCAC <u>A</u> CAACTGAG	5755
	CTCAGTTG <u>T</u> GTGAATAG	5756
Lys Overproduction DHPS 1 Triticum aestivum Ser 1654sn	TCATCGGGCATACTGTTAACTGCTTTGGAGCCAACATTAAAGTGAT AGGCAACACGGGAAATAACTCAACCAGAGAAGCTGTTCACGCGA CAGAGCAGGGATTTGCTGTTGGCATGCATGC	5757

Ser165Asn AGT-AAT

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Alteration	GCATGCATGCCAACAGCAAATCCCTGCTCTGTCGCGTGAACAGCT TCTCTGGTTGAGTTATITCCCGTGTTGCCTATCACTTTAATGTTGG CTCCAAAGCAGTTAACAGTATGCCCGATGA	5758
	CACGGGAAATAACTCAA	5759
	TTGAGTTATITCCCGTG	5760
Lys Overproduction DHPS 1	GAGCCAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACCA GAGAAGCTGTTCACGTGACAGAGCAGGGATTTGCTGTTGGCATG	5761
<i>Triticum aestivum</i> Ala174Val GCG-GTG	CATGCAGCTCTTCATGTCAATCCTTACTACGG CCGTAGTAAGGATTGACATGAAGAGCTGCATGCCAACAGCA AATCCCTGCTCTGTCACGTGAACAGCTTCTCTGGTTGAGTTACTT CCCGTGTTGCCTATCACTTTAATGTTGGCTC	5762
·	TGTTCACGTGACAGAGC	5763
·	GCTCTGTCACGTGAACA	5764
Lys Overproduction DHPS 1	GGAGCCAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACC AGAGAAGCTGTTCACACGACAGAGCAGGGATTTGCTGTTGGCAT GCATGCAGCTCTTCATGTCAATCCTTACTACG	5765
<i>Triticum aestivum</i> Ala174Thr GCG-ACG	CGTAGTAAGGATTGATGATGATGATGATGATGATGATGATG	5766
	CTGTTCACACGACAGAG	5767
	CTCTGTCGTGTGAACAG	5768
Lys Overproduction DHPS 2	TCATCGGGCACACTGTTAACTGCTTTGGAACTAACATTAAAGTGAT AGGCAACACGGGAAATAACTCAACTAGAGAAGCGATTCACGCTTC AGAGCAGGGATTTGCTGTTGGCATGCATGC	5769
Triticum aestivum Ser154Asn AGT-AAT	GCATGCACGCAACAGCAAATCCCTGCTCTGAAGCGTGAATCGCT TCTCTAGTTGAGTTA <u>T</u> TTCCCGTGTTGCCTATCACTTTAATGTTAGT TCCAAAGCAGTTAACAGTGTGCCCGATGA	5770
	CACGGGAAATAACTCAA	577
-	TTGAGTTATTTCCCGTG	577
Lys Overproduction DHPS 2 Triticum aestivum	GAACTAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACTA GAGAAGCGATTCACGTTTCAGAGCAGGGATTTGCTGTTGGCATGC	1
Ala163Val GCT-GTT	CCATAGTAAGGATTGACATGGAGAGCTGCATGCCAACAGCA AATCCCTGCTCTGAAACGTGAATCGCTTCTCTAGTTGAGTTACTTC CCGTGTTGCCTATCACTTTAATGTTAGTTC	
	GATTCACG_TTCAGAGC	577
	GCTCTGAAACGTGAATC	577

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Phenotype, Gene, Plant & Targeted Afteration	Altering Oligos	SEQ ID NO:
Lys Overproduction DHPS 2 <i>Triticum aestivum</i>	GGAACTAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACT AGAGAAGCGATTCACACTTCAGAGCAGGGATTTGCTGTTGGCATG CATGCAGCTCTCCATGTCAATCCTTACTATG	5777
Ala163Thr GCT-ACT	CATAGTAAGGATTGACATGGAGAGCTGCATGCCAACAGCAA ATCCCTGCTCTGAAG <u>T</u> GTGAATCGCTTCTCTAGTTGAGTTACTTCC CGTGTTGCCTATCACTTTAATGTTAGTTCC	5778
	CGATTCACACTTCAGAG	5779
	CTCTGAAG <u>T</u> GTGAATCG	5780
Lys Overproduction DHPS Coix lacryma-jobi	CTCATTGGGCATACTGTGAACTGCTTTGGCTCTAGAATTAAAGTGA TAGGCAACACAGGAA <u>A</u> TAACTCAACCAGAGAAGCTGTTCACGCAA CAGAGCAGGGATTTGCTGTTGGCATGCATG	5781
Ser154Asn AGT-AAT	CATGCATGCCAACAGCAAATCCCTGCTCTGTTGCGTGAACAGCTT CTCTGGTTGAGTTA <u>T</u> TTCCTGTGTTGCCTATCACTTTAATTCTAGA GCCAAAGCAGTTCACAGTATGCCCCAATGAG	5782
	CACAGGAA <u>A</u> TAACTCAA	5783
	TTGAGTTATTTCCTGTG	5784
Lys Overproduction DHPS Coix lacryma-jobi	GCTCTAGAATTAAAGTGATAGGCAACACAGGAAGTAACTCAACCA GAGAAGCTGTTCACG <u>T</u> AACAGAGCAGGGATTTGCTGTTGGCATGC ATGCAGCTCTCCACATCAATCCTTACTATGG	5785
Ala163Val GCA-GTA	CCATAGTAAGGATTGATGTGGAGAGCTGCATGCCAACAGCA AATCCCTGCTCTGTT <u>A</u> CGTGAACAGCTTCTCTGGTTGAGTTACTTC CTGTGTTGCCTATCACTTTAATTCTAGAGC	5786
	TGTTCACG <u>T</u> AACAGAGC	5787
	GCTCTGTT <u>A</u> CGTGAACA	5788
Lys Overproduction DHPS Coix lacryma-jobi	GGCTCTAGAATTAAAGTGATAGGCAACACAGGAAGTAACTCAACC AGAGAAGCTGTTCACAACAGAGCAGGGATTTGCTGTTGGCATG CATGCAGCTCTCCACATCAATCCTTACTATG	5789
Ala163Thr GCA-ACA	CATAGTAAGGATTGATGTGGAGAGCTGCATGCCAACAGCAA ATCCCTGCTCTGTTGTGTGAACAGCTTCTCTGGTTGAGTTACTTCC TGTGTTGCCTATCACTTTAATTCTAGAGCC	5790
	CTGTTCAC <u>A</u> CAACAGAG	5791
	CTCTGTTG <u>T</u> GTGAACAG	5792
Lys Overproduction DHPS Nicotiana tabacum	TCATTGGTCACACAGTCAATTGTTTTGGAGGGTCCATCAAAGTCAT CGGGAACACTGGAAACACTCCACAAGGGAAGCAATCCATGCAA CTGAACAGGGATTTGCTGTAGGTATGCATGC	5793
Ser136Asn AGC-AAC	GCATGCATACCTACAGCAAATCCCTGTTCAGTTGCATGGATTGCTT CCCTTGTGGAGTTGTTTCCAGTGTTCCCGATGACTTTGATGGACC CTCCAAAACAATTGACTGTGTGACCAATGA	5794
	CACTGGAAACACTCCA	5795

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGGAGTTGTTTCCAGTG	5796
Lys Overproduction DHPS <i>Nicotiana tabacum</i> Ala145Val GCA-GTA	GAGGGTCCATCAAAGTCATCGGGAACACTGGAAGCAACTCCACAA GGGAAGCAATCCATGTAACTGAACAGGGATTTGCTGTAGGTATGC ATGCAGCTCTTCACATTAATCCCTACTATGG	5797
	CCATAGTAGGGATTAATGCGTAGTATGC CCATAGTAGGGATTAATGTGAAGAGCTGCATGCATACCTACAGCA AATCCCTGTTCAGTTACATGGATTGCTTCCCTTGTGGAGTTGCTTC CAGTGTTCCCGATGACTTTGATGGACCCTC	5798
	AATCCATGTAACTGAAC	5799
		5800
Lys Overproduction DHPS	GTTCAGTTACATGATTACCCTACTATG	5801
Nicotiana tabacum Ala145Thr GCA-ACA	GCATGCAGCTCTTCACATTAATCCCTACTATG CATAGTAGGGATTAATGTGAAGAGCTGCATGCATACCTACAGCAA ATCCCTGTTCAGTTGTATGGATTGCTTCCCTTGTGGAGTTGCTTCC AGTGTTCCCGATGACTTTGATGGACCCTCC	5802
	CAATCCATACAACTGAA	5803
	TTCAGTTGTATGGATTG	5804
Lys Overproduction DHPS	TTATAGGCCATACCGTTAACTGTTTTGGCGGAAGCATCAAAGTCAT TGGAAACACTGGAAACAATTCGACTAGAGAAGCAATCCACGCGAC TGAACAAGGATTCGCGGTTGGAATGCATGC	5805
Arabidopsis thaliana Ser142Asn AGC-AAC	GCATGCATTCCAACCGCGAATCCTTGTTCAGTCGCGTGGATTGCT TCTCTAGTCGAATTGTTTCCAGTGTTTCCAATGACTTTGATGCTTC CGCCAAAACAGTTAACGGTATGGCCTATAA	5806
	CACTGGAAACAATTCGA	5807
	TCGAATTG <u>T</u> TTCCAGTG	5808
Lys Overproduction DHPS Arabidopsis thaliana Ala151Val GCG-GTG	GCGGAAGCATCAAAGTCATTGGAAACACTGGAAGCAATTCGACTA GAGAAGCAATCCACGTGACTGAACAAGGATTCGCGGTTGGAATGC ATGCTGCTCTTCATATAAACCCTTACTATGG	5809
	CCATAGTAAGGGTTTATATGAAGAGCAGCATGCATTCCAACCGCG AATCCTTGTTCAGTCACGTGGATTGCTTCTAGTCGAATTGCTTC CAGTGTTTCCAATGACTTTGATGCTTCCGC	581
	AATCCACG <u>T</u> GACTGAAC	581
	GTTCAGTC A CGTGGATT	581
Lys Overproduction DHPS Archidensis thaliana	GGCGGAAGCATCAAAGTCATTGGAAACACTGGAAGCAATTCGACT AGAGAAGCAATCCACACGACTGAACAAGGATTCGCGGTTGGAATG	
Arabidopsis thaliana Ala151Thr GCG-ACG	CATAGTAAGGGTTTATATGAAGAGCAGCATGCATTCCAACCGCGA ATCCTTGTTCAGTCGTGTGGATTGCTTCTCTAGTCGAATTGCTTCC AGTGTTTCCAATGACTTTGATGCTTCCGCC	581

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CAATCCAC <u>A</u> CGACTGAA	5815
	TTCAGTCGTGTGGATTG	5816
Lys Overproduction DHPS Glycine max	TTATTGCTCATACAGTCAACTGTTTTGGTGGGAAAATTAAGGTTATT GGAAATACTGGAAACAACTCCACCAGGGAAGCAATTCATGCCACT GAGCAGGGTTTTGCTGTTGGAATGCATGC	5817
Ser103Asn AGC-AAC	GCATGCATTCCAACAGCAAAACCCTGCTCAGTGGCATGAATTGCT TCCCTGGTGGAGTTG <u>T</u> TTCCAGTATTTCCAATAACCTTAATTTTCC CACCAAAACAGTTGACTGTATGAGCAATAA	5818
•	TACTGGAA <u>A</u> CAACTCCA	5819
	TGGAGTTG <u>T</u> TTCCAGTA	5820
Lys Overproduction DHPS Glycine max	GTGGGAAAATTAAGGTTATTGGAAATACTGGAAGCAACTCCACCA GGGAAGCAATTCATGTCACTGAGCAGGGTTTTGCTGTTGGAATGC ATGCTGCCCTTCACATAAACCCTTACTATGG	5821
Ala112Val GCC-GTC	CCATAGTAAGGGTTTATGTGAAGGGCAGCATGCATTCCAACAGCA AAACCCTGCTCAGTGACATGAATTGCTTCCCTGGTGGAGTTGCTT CCAGTATTTCCAATAACCTTAATTTTCCCAC	5822
	AATTCATG <u>T</u> CACTGAGC	5823
	GCTCAGTG <u>A</u> CATGAATT	5824
Lys Overproduction DHPS Glycine max	GGTGGGAAAATTAAGGTTATTGGAAATACTGGAAGCAACTCCACC AGGGAAGCAATTCATACCACTGAGCAGGGTTTTGCTGTTGGAATG CATGCTGCCCTTCACATAAACCCTTACTATG	5825
Ala112Thr GCC-ACC	CATAGTAAGGGTTTATGTGAAGGGCAGCATGCATTCCAACAGCAA AACCCTGCTCAGTGGTATGAATTGCTTCCCTGGTGGAGTTGCTTC CAGTATTTCCAATAACCTTAATTTTCCCACC	5826
	CAATTCAT <u>A</u> CCACTGAG	5827
	CTCAGTGG <u>T</u> ATGAATTG	5828
Trp Overproduction AS Arabidopsis thaliana Asp341Asn GAC-AAC	CTTGCAGGAGACATATTTCAGATCGTGCTGAGTCAACGTTTTGAG CGGCGAACATTTGCAAACCCCTTTGAAGTTTATAGAGCACTAAGA GTTGTGAATCCAAGTCCGTATATGGGTTATT	5829
	AATAACCCATATACGGACTTGGATTCACAACTCTTAGTGCTCTATA AACTTCAAAGGGGTTTGCAAATGTTCGCCGCTCAAAACGTTGACT CAGCACGATCTGAAATATGTCTCCTGCAAG	5830
	CATTTGCA <u>A</u> ACCCCTTT	5831
	AAAGGGGT <u>T</u> TGCAAATG	5832
Trp Overproduction AS Nicotiana tabacum	GCTGCAGGAGACATATTTCAAATCGTTTTAAGTCAACGCTTTGAGA GAAGAACATTTGCTAACCCATTTGAAGTGTACAGAGCATTAAGAAT TGTGAATCCAAGCCCATATATGACTTACA	5833

Asp326Asn GAC-AAC

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
AII GAAAAAA	TGTAAGTCATATATGGGCTTGGATTCACAATTCTTAATGCTCTGTA CACTTCAAATGGGTTAGCAAATGTTCTTCTCTCAAAGCGTTGACTT AAAACGATTTGAAATATGTCTCCTGCAGC	5834
	CATTIGCTAACCCATIT	5835
	AAATGGGT <u>T</u> AGCAAATG	5836
Trp Overproduction AS Oryza sativa Asp323Asn GAC-AAC	CTAGCTGGTGACATTTTCAAGTAGTCTTAAGCCAGCGTTTTGAGA GGCGTACATTTGCTAACCCCTTTGAGGTGTACCGTGCATTGCGTA TTGTCAATCCTAGTCCTTATATGGCCTATC	5837
	GATAGGCCATATAGGGATTGACAATACGCAATGCACGGT ACACCTCAAAGGGGTTAGCAAATGTACGCCTCTCAAAACGCTGGC TTAAGACTACTTGAAAAATGTCACCAGCTAG	5838
	CATTTGCTAACCCCTTT	5839
	AAAGGGTTAGCAAATG	5840
Trp Overproduction AS Ruta graveolens Asp354Asn GAC-AAC	CTTGCTGGTGACATATTCCAGATCGTACTAAGTCAGCGTTTTGAAA GGCGAACGTTCGCAAACCCATTTGAAATCTATAGATCACTGAGGA	5841
	TTGTTAATCCAAGCCCATATATGACTTATT AATAAGTCATATATGGGCTTGGATTAACAATCCTCAGTGATCTATA GATTTCAAATGGGTTTGCGAACGTTCGCCTTTCAAAACGCTGACTT AGTACGATCTGGAATATGTCACCAGCAAG	5842
	CGTTCGCAAACCCATTT	5843
	AAATGGGT <u>T</u> TGCGAACG	584
Trp Overproduction AS Catharanthus roseus Asp354Asn GAT-AAT	CTGGCTGGGACATATTCCAGCTTGTCCTAAGTCAGCGTTTTGAA CGGCGAACATTTGCAAATCCATTTGAAGTCTACCGAGCATTGAGA ATTGTCAACCCAAGTCCATATATGACTTATT	584
	AATAAGTCATATATGGACTTGGGTTGACAATTCTCAATGCTCGGTA GACTTCAAATGGATTTGCAAATGTTCGCCGTTCAAAACGCTGACTT AGGACAAGCTGGAATATGTCCCCAGCCAG	584
	CATTIGCAAATCCATTI	584
	AAATGGATTTGCAAATG	584

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Example 10

Production of modified starch in plants

A principal aim of biotechnology is the improvement of crop plants for food value, agriculture, and to produce a range of plant-derived raw materials. Along with oils, fats and proteins, polysaccharides constitute the main raw materials derived from plants, and apart from cellulose, the storage polymer starch is the most important polysaccharide raw material. Starch is derived from a range of plants, but maize is the most important cultivated plant for the production of starch.

The polysaccharide starch is a polymer made up of glucose molecules. However, starch is not a homogeneous raw material and is, in fact, a highly complex mixture of various types of molecules which differ from each other, for example, in their degree of polymerization and in the degree of branching of the glucose chains. For example, amylose-starch is a basically non-branched polymer made up of α -1,4-glycosidically branched glucose molecules, and amylopectin-starch is a complex mixture of variously branched glucose chains. The branching results from additional α -1,6-glycosidic linkages. In plants from which starch is typically isolated, for example maize or potato, the starch is approximately 25% amylose-starch and 75% amylopectin-starch.

In maize, various mutants in starch metabolism are known, for example waxy, sugary, shrunken and opaque-2. In addition to producing a modified starch, these mutations greatly improve grain quality in maize, and thus expand the use of maize not only as the food but also for the important industrial materials in food chemistry. It would therefore be advantageous to be able readily to obtain mutants in these genes in particular maize genotypes as well as other plants. Such plants can be obtained, for example, using traditional breeding methods and through specific genetic modification by means of recombinant DNA techniques.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations in genes involved in starch metabolism.

Table 20
Genome-Altering Oligos Conferring Increased Starch

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
ncreased Starch ADPGPP	GAACTTGAGACTGAGAAAAGGGATCCAAGGACAGTTGCTTCCATT ATTCTTGGAGGTGGAAAAGGGAACTCGACTCTTTCCTCTCACAAAA CGCCGCGCCAAGCCTGCCGTTCCTATCGGGG	5849
Arabidopsis thaliana Ala99Lys GCA-AAA	CCCGATAGGAACGCAGGCTTGGCGCGCGTTTTGTGAGAGGA AAGAGTCGAGTTCCT <u>TT</u> TCCACCTCCAAGAATAATGGAAGCAACT GTCCTTGGATCCCTTTTCTCAGTCTCAAGTTC	5850
•	GAGGTGGA <u>AA</u> AGGAACT	5851
•	AGTTCCT <u>TT</u> TCCACCTC	5852
Increased Starch ADPGPP Arabidopsis thaliana	CAAAACGCCGCGCCAAGCCTGCCGTTCCTATCGGGGGAGCCTAT AGGTTGATAGATGTACTAATGAGCAATTGTATTAACAGCGGAATCA ACAAAGTCTACATACTCACACAATATAACTC	5853
Pro127Leu CCA-CTA	GAGTTATATTGTGTGAGTATGTAGACTTTGTTGATTCCGCTGTTAA TACAATTGCTCATTAGTACATCTATCAACCTATAGGCTCCCCCGAT AGGAACGGCAGGCTTGGCGCGCGCGTTTTG	5854
	AGATGTACTAATGAGCA	5855
	TGCTCATTAGTACATCT	5856
Increased Starch ADPGPP	TCACACAATATAACTCAGCATCATTGAACAGGCATTTAGCCCGTGC TTACAACTCCAAT <u>AAT</u> CTTGGCTTTGGAGATGGCTATGTTGAGGTT CTTGCGGCCACTCAAACGCCAGGAGAATC	5857
Arabidopsis thaliana Gly162Asn GGA-AAT	GATTCTCCTGGCGTTTGAGTGGCCGCAAGAACCTCAACATAGCCA TCTCCAAAGCCAAG <u>ATT</u> ATTGGAGTTGTAAGCACGGGCTAAATGC CTGTTCAATGATGCTGAGTTATATTGTGTGA	585
	CTCCAAT <u>AAT</u> CTTGGCT	585
	AGCCAAGATTATTGGAG	586
Increased Starch ADPGPP Arabidopsis thaliana Gly162Asn GGA-AAC	TCACACAATATAACTCAGCATCATTGAACAGGCATTTAGCCCGTGC TTACAACTCCAATAACCTTGGCTTTGGAGATGGCTATGTTGAGGTT CTTGCGGCCACTCAAACGCCAGGAGAATC	586
	GATTCTCCTGGCGTTTGAGTGGCCGCAAGAACCTCAACATAGCCA TCTCCAAAGCCAAGGTTATTGGAGTTGTAAGCACGGGCTAAATGC CTGTTCAATGATGCTGAGTTATATTGTGTGA	586
	CTCCAATAACCTTGGCT	586
	AGCCAAGGTTATTGGAG	586

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Increased Starch ADPGPP Arabidopsis thaliana Asn100Lys AAT-AAA	GTTTGAGAGAAGAAAGGTAGACCCGCAAAATGTGGCTGCAATCAT TCTAGGAGGAGGCAAAGGAGCTAAACTCTTCCCTCTTACAATGAG AGCCGCAACACCAGCTGTAAATATTCATCTT	5865
	AAGATGAATATTTACAGCTGGTGTTGCGGCTCTCATTGTAAGAGG GAAGAGTTTAGCTCCTTTGCCTCCTCCTAGAATGATTGCAGCCAC ATTTTGCGGGTCTACCTTTCTTCTCTCAAAC	5866
	GGAGGCAA <u>A</u> GGAGCTAA	5867
	TTAGCTCCTTTGCCTCC	5868
Increased Starch ADPGPP Arabidopsis thaliana	CTTGTGTCTTCAAATTATGTTAGGTTCCTGTTGGTGGATGCTACAG GCTGATCGATATCC <u>T</u> GATGAGTAACTGTATTAACAGCTGCATCAAC AAGATATTTGTGCTGACACAGTTCAACTC	5869
Pro128Leu CCG-CTG	GAGTTGAACTGTGTCAGCACAAATATCTTGTTGATGCAGCTGTTAA TACAGTTACTCATCAGGATATCGATCAGCCTGTAGCATCCACCAA CAGGAACCTAACATAATTTGAAGACACAAG	5870
	CGATATCC <u>T</u> GATGAGTA	5871
	TACTCATC <u>A</u> GGATATCG	5872
Increased Starch ADPGPP Arabidopsis thaliana	TGACACAGTTCAACTCAGCTTCCCTTAATCGACATTTAGCACGAAC TTATTTTGGGAATAATATAAACTTTGGAGGTGGTTTCGTAGAGGTA CAAACACTATGACAATAATAACTCTCAGC	5873
Gly163Asn GGC-AAT	GCTGAGAGTTATTGTCATAGTGTTTGTACCTCTACGAAACCAC CTCCAAAGTTTATATTATTCCCAAAATAAGTTCGTGCTAAATGTCG ATTAAGGGAAGCTGAGTTGAACTGTGTCA	5874
	TGGGAAT <u>AAT</u> ATAAACT	5875
	AGTTTAT <u>ATT</u> ATTCCCA	5876
Increased Starch ADPGPP Arabidopsis thaliana	TGACACAGTTCAACTCAGCTTCCCTTAATCGACATTTAGCACGAAC TTATTTTGGGAATAACACTTTGGAGGTGGTTTCGTAGAGGTA CAAACACTATGACAATAATAACTCTCAGC	5877
Gly163Asn GGC-AAC	GCTGAGAGTTATTATTGTCATAGTGTTTGTACCTCTACGAAACCAC CTCCAAAGTTTAT <u>GTT</u> ATTCCCAAAATAAGTTCGTGCTAAATGTCG ATTAAGGGAAGCTGAGTTGAACTGTGTCA	5878
	TGGGAAT <u>AAC</u> ATAAACT	5879
	AGTITAT <u>GTT</u> ATTCCCA	5880
Increased Starch ADPGPP Lycopersicon	TTGAGGAACAACCAACGGCAGATCCAAAAGCTGTTGCCTCTGTCA TTCTAGGTGGTGGTAAAGGAACTCGTCTTTTTCCTCTTACAAGCA GAAGAGCTAAACCAGCTGTTCCTATTGGTGG	5881
esculentum Val94Lys GTT-AAA	CCACCAATAGGAACAGCTGGTTTAGCTCTTCTGCTTGTAAGAGGA AAAAGACGAGTTCC <u>TTT</u> ACCACCACCTAGAATGACAGAGGCAACA GCTTTTGGATCTGCCGTTGGTTGTTCCTCAA	5882
	TGGTGGT <u>AAA</u> GGAACTC	5883

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	GAGTTCC <u>TTT</u> ACCACCA	5884
ncreased Starch	CAAGCAGAAGAGCTAAACCAGCTGTTCCTATTGGTGGTTGTTACC GGCTAATTGATGTACAAATGAGTAACTGCATTAACAGTGGCATAC	5885
Lycopersicon esculentum Pro122Leu	GGAAAATTITCATCTTAACACAGTTCAATTC GAATTGAACTGTGTTAAGATGAAAATTTTCCGTATGCCACTGTTAA TGCAGTTACTCATTTGTACATCAATTAGCCGGTAACAACCACCAAT AGGAACAGCTGGTTTAGCTCTTCTGCTTG	5886
CCA-CAA	TGATGTACAAATGAGTA	5887
	TACTCATT <u>T</u> GTACATCA	5888
Increased Starch ADPGPP	CACAGTTCAATTCCTTTTCCCTCAATCGTCACCTTGCCCGCACGTA TAATTTTGGAAATATGTGGGTTTTGGAGATGGATTTGTGGAGGTT TTAGCTGCAACCCAGACTCCAGGGGATGC	5889
Lycopersicon esculentum Gly158Asn	GCATCCCTGGAGTCTGGGTTGCAGCTAAAACCTCCACAAATCCA TCTCCAAAACCCACATTATTTCCAAAATTATACGTGCGGGCAAGGT	5890
GGA-AAT	GACGATTGAGGGAAAAGGAATTGAACTGTG TGGAAAT AAT GTGGGTT	589
	AACCCAC <u>ATT</u> ATTTCCA	589
Increased Starch ADPGPP	CACAGTTCAATTCCTTTTCCCTCAATCGTCACCTTGCCCGCACGTA TAATTTTGGAAATAACGTGGGGTTTTGGAGATGGATTTGTGGAGGTT	589
Lycopersicon esculentum Gly158Asn	TTAGCTGCAACCCAGACTCCAGGGGATGC GCATCCCCTGGAGTCTGGGTTGCAGCTAAAACCTCCACAAATCCA TCTCCAAAACCCACGTTATTTCCAAAATTATACGTGCGGGCAAGGT GACGATTGAGGGAAAAGGAATTGAACTGTG	589
GGA-AAC	TGGAAAT <u>AAC</u> GTGGGTT	589
·	AACCCACGTTATTTCCA	589
Increased Starch ADPGPP Cicer arietinum Ala101Lys GCT-AAA	ACGTAGATTTGGAAAAAAGAGACCCAAGTACAGTTGTAGCAATTAT ACTAGGTGGAGGT <u>AAA</u> GGAACTCGTCTCTTCCCTCTCACCAAGC GACGAGCCAAGCCTGCTGTTCCAATTGGAGG	589
	CCTCCAATTGGAACAGCAGGCTTGGCTCGCTTGGTGAGAGG GAAGAGACGAGTTCC <u>TTT</u> ACCTCCACCTAGTATAATTGCTACAACT GTACTTGGGTCTCTTTTTTCCAAATCTACGT	
	TGGAGGTAAAGGAACTC	58
	GAGTTCCTTTACCTCCA	59

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Increased Starch ADPGPP Cicer arietinum Pro129Leu CCA-CTA	CCAAGCGACGAGCCAAGCCTGCTGTTCCAATTGGAGGTGCTTATA GGCTGATAGATGTACTAATGAGTAACTGCATCAATAGTGGGATCAA CAAAGTATACATTCTCACTCAATTTAATTC	5901
	GAATTAAATTGAGTGAGAATGTATACTTTGTTGATCCCACTATTGAT GCAGTTACTCATTAGTACATCTATCAGCCTATAAGCACCTCCAATT GGAACAGCAGGCTTGGCTCGTCGCTTGG	5902
	AGATGTAC <u>T</u> AATGAGTA	5903
	TACTCATT <u>A</u> GTACATCT	5904
Increased Starch ADPGPP Cicer arietinum	CTCAATTTAATTCAGCCTCACTCAACAGGCATATTGCACGTGCTTA TAACTCTGGTACT AAT GTCACTTTTGGAGATGGCTATGTTGAGGTT CTTGCAGCAACTCAAACTCCAGGGGAGCA	5905
Gly165Asn GGA-AAT	TGCTCCCTGGAGTTTGAGTTGCTGCAAGAACCTCAACATAGCCA TCTCCAAAAGTGACATTAGTACCAGAGTTATAAGCACGTGCAATAT GCCTGTTGAGTGAGGCTGAATTAAATTGAG	5906
	TGGTACT <u>AAT</u> GTCACTT	5907
_	AAGTGAC <u>ATT</u> AGTACCA	5908
Increased Starch ADPGPP Cicer arietinum	CTCAATTTAATTCAGCCTCACTCAACAGGCATATTGCACGTGCTTA TAACTCTGGTACT AAC GTCACTTTTGGAGATGGCTATGTTGAGGTT CTTGCAGCAACTCAAACTCCAGGGGAGCA	5909
Gly165Asn GGA-AAC	TGCTCCCTGGAGTTTGAGTTGCTGCAAGAACCTCAACATAGCCA TCTCCAAAAGTGACGTTAGTACCAGAGTTATAAGCACGTGCAATAT GCCTGTTGAGTGAGGCTGAATTAAATTGAG	5910
	TGGTACT AAC GTCACTT	5911
	AAGTGAC GTT AGTACCA	5912
Increased Starch ADPGPP Ipomoea batatas Ala94Lys GCA-AAA	ATATTGGAGAGGCGTCGGGCAAACCCTAAGAATGTGGCTGCAATC ATACTGCCAGGCGGTAAAGGGGACACACCTATTCCCTCTCACCAAT CGAGCTGCAACCCCTGCTGTTCCACTTGGAG	5913
	CTCCAAGTGGAACAGCAGGGGTTGCAGCTCGATTGGTGAGAGGG AATAGGTGTCCCTTTACCGCCTGGCAGTATGATTGCAGCCACA TTCTTAGGGTTTGCCCGACGCCTCTCCAATAT	5914
	CAGGCGGT <u>AA</u> AGGGACA	5915
	TGTCCCTTTACCGCCTG	5916
Increased Starch ADPGPP Ipomoea batatas	CCAATCGAGCTGCAACCCCTGCTGTTCCACTTGGAGGATGCTATA GGTTGATCGACATTCTAATGAGCAACTGCATCAACAGCGGGGTTA ACAAGATCTTTGTGCTGACCCAGTTCAATTC	5917
Pro122Leu CCA-CTA	GAATTGAACTGGGTCAGCACAAAGATCTTGTTAACCCCGCTGTTG ATGCAGTTGCTCATTAGAATGTCGATCAACCTATAGCATCCTCCAA GTGGAACAGCAGGGGTTGCAGCTCGATTGG	5918
	CGACATTC <u>T</u> AATGAGCA	5919

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Aicounon	TGCTCATT A GAATGTCG	5920
Increased Starch ADPGPP	TGACCCAGTTCAATTCAGCTTCTCTTAACCGTCACATTTCCCGTAC CGTCTTTGGCAAT <u>AAT</u> GTGAGCTTCGGAGATGGATTTGTTGAGGT GCTGGCTGCAACCCAAACACAAGGGGAAAC	5921
pomoea batatas Gly157Asn GGT-AAT	GTTTCCCCTTGTGTTTGGGTTGCAGCCAGCACCTCAACAAATCCA TCTCCGAAGCTCACATTATTGCCAAAGACGGTACGGGAAATGTGA CGGTTAAGAGAAGCTGAATTGAACTGGGTCA	5922
	TGGCAAT AAT GTGAGCT	5923
	AGCTCACATTATTGCCA	5924
Increased Starch ADPGPP	TGACCCAGTTCAATTCAGCTTCTCTTAACCGTCACATTTCCCGTAC CGTCTTTGGCAATAACGTGAGCTTCGGAGATGGATTTGTTGAGGT GCTGGCTGCAACCCAAACACAAGGGGAAAC	5925
<i>lpomoea batatas</i> Gly157Asn GGT-AAC	GTTTCCCTTGTGTTTGGGTTGCAGCCAGCACCTCAACAAATCCA TCTCCGAAGCTCACGTTATTGCCAAAGACGGTACGGGAAATGTGA CGGTTAAGAGAAGCTGAATTGAACTGGGTCA	5926
	TGGCAATAACGTGAGCT	5927
	AGCTCAC <u>GTT</u> ATTGCCA	5928
Increased Starch ADPGPP	CATTCCGGAGGAACTTTGCGGATCCAAATGAGGTTGCTGCTGTTA TATTGGGTGGTGGCAAAGGGACTCAACTTTTTCCTCTCACAAGCA CAAGGGCCACGCCTGCTGTTCCTATTGGAGG	5929
Oryza sativa Thr96Lys ACC-AAA	CCTCCAATAGGAACAGCAGGCGTGGCCCTTGTGCTTGTGAGAGG AAAAAGTTGAGTCCCTTTGCCACCACCCAATATAACAGCAGCAAC CTCATTTGGATCCGCAAAGTTCCTCCGGAATG	5930
	TGGTGGCA <u>AA</u> GGGACTC	593′
	GAGTCCCTTTGCCACCA	593
Increased Starch ADPGPP	CAAGCACAAGGGCCACGCCTGCTGTTCCTATTGGAGGATGCTATA GGCTTATCGATATCCTCATGAGCAACTGTTTCAACAGTGGCATAAA CAAGATATTCATAATGACTCAATTCAACTC	593
Oryza sativa Pro1.24Leu CCC-CTC	GAGTTGAATTGAGTCATTATGAATATCTTGTTTATGCCACTGTTGAA ACAGTTGCTCATGAGGATATCGATAAGCCTATAGCATCCTCCAATA GGAACAGCAGGCGTGGCCCTTGTGCTTG	593
	CGATATCCTCATGAGCA	593
	TGCTCATGAGGATATCG	593
Increased Starch ADPGPP	TGACTCAATTCAACTCAGCATCTCTTAATCGTCACATTCATCGTAC GTACCTTGGTGGTAATATCAACTTTACTGATGGTTCTGTTGAGGTA TTAGCCGCTACACAAATGCCTGGGGAAGGC	593
Oryza sativa Gly159Asn GGA-AAT	GCTCCCCAGGCATTTGTGTAGCGGCTAATACCTCAACAGAACCA TCAGTAAAGTTGATATTACCACCAAGGTACGATGAATGTGAC GATTAAGAGATGCTGAGTTGAATTGAGTCA	593

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGGTGGTAATATCAACT	5939
	AGTTGAT <u>ATT</u> ACCACCA	5940
Increased Starch ADPGPP Oryza sativa	TGACTCAATTCAACTCAGCATCTCTTAATCGTCACATTCATCGTAC GTACCTTGGTGGTAACATCAACTTTACTGATGGTTCTGTTGAGGTA TTAGCCGCTACACAAATGCCTGGGGAGGC	5941
Gly159Asn GGA-AAC	GCCTCCCAGGCATTTGTGTAGCGGCTAATACCTCAACAGAACCA TCAGTAAAGTTGAT <u>GTT</u> ACCACCAAGGTACGTACGATGAATGTGA CGATTAAGAGATGCTGAGTTGAATTGAGTCA	5942
	TGGTGGT <u>AAC</u> ATCAACT	5943
	AGTTGAT <u>GTT</u> ACCACCA	5944
Increased Starch ADPGPP Triticum aestivum	GTCCTTCAGGAGGATTAAGCGATCCGAACGAGGTTGCGGCCGTC ATACTCGGCGGCGGCA <u>AA</u> GGGACTCAGCTCTTCCCACTCACGAG CACAAGGGCCACACCTGCTGTTCCTATTGGAGG	5945
Thr80Lys ACC-AAA	CCTCCAATAGGAACAGCAGGTGTGGCCCTTGTGCTCGTGAGTGG GAAGAGCTGAGTCCC <u>TT</u> TGCCGCCGCCGAGTATGACGGCCGCAA CCTCGTTCGGATCGCTTAATCCTCCTGAAGGAC	5946
	CGGCGGCA <u>AA</u> GGGACTC	5947
ļ	GAGTCCC <u>TT</u> TGCCGCCG	5948
Increased Starch ADPGPP Triticum aestivum	CGAGCACAAGGGCCACACCTGCTGTTCCTATTGGAGGATGTTACA GGCTCATCGACATTCTCATGAGCAACTGCTTCAACAGTGGCATCA ACAAGATATTCGTCATGACCCAGTTCAACTC	5949
Pro108Leu CCC-CTC	GAGTTGAACTGGGTCATGACGAATATCTTGTTGATGCCACTGTTG AAGCAGTTGCTCATGAGAATGTCGATGAGCCTGTAACATCCTCCA ATAGGAACAGCAGGTGTGGCCCTTGTGCTCG	5950
	CGACATTC <u>T</u> CATGAGCA	5951
	TGCTCATGAGAATGTCG	5952
Increased Starch ADPGPP Triticum aestivum	TGACCCAGTTCAACTCGGCCTCCCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGGAATATCAATTTCACTGATGGATCCGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5953
Gly143Asn GGA-AAT	GCCTCCCGGGCATTTGCGTCGCGGCCAATACCTCAACGGATCC ATCAGTGAAATTGAT <u>ATT</u> CCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGGGAGGCCGAGTTGAACTGGGTCA	5954
	CGGCGGG <u>AAT</u> ATCAATT	5955
	AATTGAT <u>ATT</u> CCCGCCG	5956
Increased Starch ADPGPP Triticum aestivum Glv143Asn	TGACCCAGTTCAACTCGGCCTCCCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGGAACATCAATTTCACTGATGGATCCGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5957

Gly143Asn GGA-AAC

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	GCCTCCCGGGCATTTGCGTCGCGGCCAATACCTCAACGGATCC ATCAGTGAAATTGATGTTCCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGGGAGGCCGAGTTGAACTGGGTCA	5958
•	CGCCGGAACATCAATT	5959
	AATTGAT <u>GTT</u> CCCGCCG	5960
ncreased Starch	CCTCCCGAAAGAATTATGCTGATGCAAGCCACGTTTCTGCTGTCA TTTTGGGTGGAGGCAAAGGAATTCAACTCTTTCCTCTGACAAGCA CAAGGGCTACCCCCGCTGTTCCTGTTGGAGG	5961
O <i>ryza sativa</i> Thr95Lys ACT-AAA	CAAGGGCTACCCCGGTGTTGTGTGTGTGTGTGTGTGAGAGG CCTCCAACAGGAACAGCGGGGGGTAGCCCTTGTGCTTGTCAGAGG AAAGAGTTGAACTCC <u>TT</u> TGCCTCCACCCAAAATGACAGCAGAAAC GTGGCTTGCATCAGCATAATTCTTTCGGGAGG	5962
	TGGAGGCA <u>AA</u> GGAGTTC	5963
	GAACTCC <u>TT</u> TGCCTCCA	5964
Increased Starch ADPGPP	CAAGCACAAGGGCTACCCCCGCTGTTCCTGTTGGAGGATGTTACA GCCTTATTGACATCCTTATGAGCAATTGCTTCAATAGCGGAATAAA	5965
Oryza sativa Pro123Leu CCT-CTT	TAAAATATTTGTGATGACTCAGTTCAATTC GAATTGAACTGAGTCATCACAAATATTTTATTT	5966
	GGAACAGCGGGGTAGCCCTTGTGCTTG TGACATCCTTATGAGCA	5967
	TGCTCATAAGGATGTCA	5968
Increased Starch ADPGPP	TGACTCATAAGGATGTCA TGACTCAGTTCAATTCTGCTTCTCTTAATCGCCATATCCATCATAC ATACCTTGGTGGGAATATCAACTTTACTGATGGGTCTGTGCAGGT ATTGGCTGCTACACAAATGCCTGACGAACC	5969
<i>Oryza sativa</i> Gly158Asn GGG-AAT	GGTTCGTCAGGCATTTGTGTAGCAGCCAATACCTGCACAGACCCA TCAGTAAAGTTGATATTCCCACCAAGGTATGTATGATGGATATGGC GATTAAGAGAAGCAGAATTGAACTGAGTCA	5970
•	TGGTGGGAATATCAACT	597
	AGTTGATATTCCCACCA	597
Increased Starch ADPGPP Oryza sativa Gly158Asn GGG-AAC	TGACTCAGTTCAATTCTGCTTCTCTTAATCGCCATATCCATCATAC ATACCTTGGTGGGAACATCAACTTTACTGATGGGTCTGTGCAGGT ATTGGCTGCTACACAAATGCCTGACGAACC	597
	GGTTCGTCAGGCATTTGTGTGTGCGCAGCCAATACCTGCACAGACCCA TCAGTAAAGTTGATGTTCCCACCAAGGTATGTATGATGGATATGGC GATTAAGAGAAGCAGAATTGAACTGAGTCA	597
	TGGTGGGAACATCAACT	597
	AGTTGATGTTCCCACCA	597

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Increased Starch ADPGPP Triticum aestivum	CCTTCCGCAGGAATTACGCCGATCCGAACGAGGTCGCGGCCGTC ATACTCGGCGGTGGCAAAGGGGACTCAGCTCTTCCCTCTCACAAG CACAAGGGCCACACCTGCTGTTCCTATTGGAGG	5977
Thr99Lys ACC-AAA	CCTCCAATAGGAACAGCAGGTGTGGCCCTTGTGCTTGTGAGAGG GAAGAGCTGAGTCCC <u>TT</u> TGCCACCGCCGAGTATGACGGCCGCGA CCTCGTTCGGATCGGCGTAATTCCTGCGGAAGG	5978
1	CGGTGGCA <u>AA</u> GGGACTC	5979
	GAGTCCC <u>TT</u> TGCCACCG	5980
Increased Starch ADPGPP Triticum aestivum	CAAGCACAAGGGCCACACCTGCTGTTCCTATTGGAGGATGTTACA GGCTCATCGATATTCTCATGAGCAACTGCTTCAATAGTGGCATCAA CAAGATATTCGTCATGACGCAGTTCAACTC	5981
Pro127Leu CCC-CTC	GAGTTGAACTGCGTCATGACGAATATCTTGTTGATGCCACTATTGA AGCAGTTGCTCATGAGAATATCGATGAGCCTGTAACATCCTCCAA TAGGAACAGCAGGTGTGGCCCTTGTGCTTG	5982
	CGATATTC <u>T</u> CATGAGCA	5983
	TGCTCATG <u>A</u> GAATATCG	5984
Increased Starch ADPGPP Triticum aestivum	TGACGCAGTTCAACTCGGCCTCTCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGGAATATCAATTTCACTGATGGATCTGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5985
Gly162Asn GGA-AAT	GCCTCCCGGGCATTTGCGTCGCGGCCAATACCTCAACAGATCC ATCAGTGAAATTGAT <u>ATT</u> CCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGAGAGGCCGAGTTGAACTGCGTCA	5986
	CGGCGGG <u>AAT</u> ATCAATT	5987
	AATTGAT <u>ATT</u> CCCGCCG	5988
Increased Starch ADPGPP Triticum aestivum	TGACGCAGTTCAACTCGGCCTCTCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGAACATCAATTTCACTGATGGATCTGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5989
Gly162Asn GGA-AAC	GCCTCCCGGGCATTTGCGTCGCGGCCAATACCTCAACAGATCC ATCAGTGAAATTGAT <u>GTT</u> CCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGAGAGGCCGAGTTGAACTGCGTCA	5990
	CGGCGGG <u>AAC</u> ATCAATT	5991
	AATTGAT GTT CCCGCCG	5992
Increased Starch ADPGPP Zea mays	CTTTTCGGAGGAATTATGCTGATCCTAATGAAGTCGCTGCCGTCA TTTTGGGTGGTGGTAAAGGGGACTCAGCTTTTCCCTCTCACAAGCA CAAGGGCCACCCCTGCTGTTCCTATTGGAGG	5993
Thr96Lys ACC-AAA	CCTCCAATAGGAACAGCAGGGGTGGCCCTTGTGCTTGTGAGAGG GAAAAGCTGAGTCCC <u>TT</u> TACCACCACCCAAAATGACGGCAGCGAC TTCATTAGGATCAGCATAATTCCTCCGAAAAG	5994
	TGGTGGTA AA GGGACTC	5995

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Alteration	GAGTCCCTTTACCACCA	5996
ncreased Starch	CAAGCACAAGGGCCACCCCTGCTGTTCCTATTGGAGGATGTTACA GGCTTATTGATATCCTCATGAGCAACTGTTTCAACAGTGGCATAAA CAAGATATTTGTTATGACTCAGTTCAACTC	5997
Zea mays Pro124Leu CCC-CTC	GAGATATTTGTTATGACTCAGTTCAACTC GAGTTGAACTGAGTCATAACAAATATCTTGTTTATGCCACTGTTGA AACAGTTGCTCATGAGGATATCAATAAGCCTGTAACATCCTCCAAT AGGAACAGCAGGGGTGGCCCTTGTGCTTG	5998
	TGATATCC <u>T</u> CATGAGCA	5999
	TGCTCATGAGGATATCA	6000
Increased Starch ADPGPP	TGACTCAGTTCAACTCAGCTTCTCTTAACCGTCACATTCATCGTAC CTATCTTGGTGGGAATATCAACTTCACTGATGGATCTGTTGAGGT GCTGGCTGCAACACAAATGCCTGGGGAGGC	6001
Zea mays Gly159Asn GGG-AAT	GCTCCCCAGGCATTTGTGTTGCAGCCAGCACCTCAACAGATCCA TCAGTGAAGTTGATATCCCACCAAGATAGGTACGATGAATGTGA CGGTTAAGAGAAGCTGAGTTGAACTGAGTCA	6002
	TGGTGGGAATATCAACT	6003
	AGTTGATATTCCCACCA	6004
Increased Starch ADPGPP	TGACTCAGTTCAACTCAGCTTCTCTTAACCGTCACATTCATCGTAC CTATCTTGGTGGGAACATCAACTTCACTGATGGATCTGTTGAGGT	600
Zea mays Gly159Asn GGG-AAC	GCTGGCTGCAACACAAATGCCTGGGGAGGC GCCTCCCCAGGCATTTGTGTTGCAGCCAGCACCTCAACAGATCCA TCAGTGAAGTTGATGTTCCCACCAAGATAGGTACGATGAATGTGA CGGTTAAGAGAAGCTGAGTTGAACTGAGTCA	600
	TGGTGGGAACATCAACT	600
	AGTTGATGTTCCCACCA	600
Increased Starch ADPGPP Solanum tuberosum	CTTGAGAGGCAAAAGAAGGGCGATGCAAGGACAGTAGTAGCAAT CATTCTAGGAGGGGGAA ACGTCGTGCTAAGCCTGCCGTTCCAATGGGAG	600
Ala58Lys GCG-AAG	CTCCCATTGGAACGGCAGGCTTAGCACGACGTTTGGTGAGGGGG AAAAGACGAGTTCCCTTTCCCCCTCCTAGAATGATTGCTACTACTG TCCTTGCATCGCCCTTCTTTTGCCTCCAAG	
	GAGGGGAAAGGGAACT	601
·	AGTTCCCTTTCCCCCTC	601
Increased Starch ADPGPP Solanum tuberosum	CCAAACGTCGTGCTAAGCCTGCCGTTCCAATGGGAGGAGCATATA GGCTAATTGATGTAC <u>T</u> AATGAGCAACTGTATTAACAGTGGCATCAA CAAAGTATACATTCTCACTCAATTCAACTC	
Pro86Leu CCA-CTA	GAGTTGAATTGAGTGAGAATGTATACTTTGTTGATGCCACTGTTAA TACAGTTGCTCATTAGTACATCAATTAGCCTATATGCTCCCCAT TGGAACGCAGGCTTAGCACGACGTTTGG	601

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGATGTAC <u>T</u> AATGAGCA	6015
	TGCTCATT <u>A</u> GTACATCA	6016
Increased Starch	CTCAATTCAACTCAGCCTCACTTAACAGGCATATAGCTCGTGCTTA	6017
ADPGPP	CAACTTTGGCAAT <u>AAT</u> GTCACATTCGAGAGTGGCTATGTCGAGGT	
Solanum tuberosum	CTTAGCAGCAACTCAAACACCAGGTGAATT	
Gly122Asn	AATTCACCTGGTGTTTGAGTTGCTGCTAAGACCTCGACATAGCCA	6018
GGG-AAT	CTCTCGAATGTGACAATTATTGCCAAAGTTGTAAGCACGAGCTATAT	
	GCCTGTTAAGTGAGGCTGAGTTGAATTGAG TGGCAAT AAT GTCACAT	6019
		6020
	ATGTGACATTATTGCCA	
Increased Starch ADPGPP	CTCAATTCAACTCAGCCTCACTTAACAGGCATATAGCTCGTGCTTA CAACTTTGGCAAT AAC GTCACATTCGAGAGTGGCTATGTCGAGGT	6021
Solanum tuberosum	CTTAGCAGCAACTCAAACACCAGGTGAATT	
Gly122Asn	AATTCACCTGGTGTTTGAGTTGCTGCTAAGACCTCGACATAGCCA	6022
GGG-AAC	CTCTCGAATGTGACGTTATTGCCAAAGTTGTAAGCACGAGCTATAT	
	GCCTGTTAAGTGAGGCTGAGTTGAATTGAG	
	TGGCAAT <u>AAC</u> GTCACAT	6023
	ATGTGAC <u>GTT</u> ATTGCCA	6024
Increased Starch	TATTTGAATCTCCAAAAGCTGACCCAAAAAATGTGGCTGCAATTGT	6025
ADPGPP	GCTGGGTGGTAAAGGGACTCGCCTCTTTCCTCTTACTAGCA	
Beta vulgaris	GGAGAGCTAAGCCAGCAGTGCCAATTGGAGG	2222
Ala98Lys GCT-AAA	CCTCCAATTGGCACTGCTGGCTTAGCTCTGCTAGTAAGAGGA	6026
GCT-AVA	AAGAGGCGAGTCCC <u>TTT</u> ACCACCACCCAGCACAATTGCAGCCACA	
	TGGTGGTAAAGGGACTC	6027
	GAGTCCCTTTACCACCA	6028
Increased Starch	TATTTGAATCTCCAAAAGCTGACCCAAAAAATGTGGCTGCAATTGT	6029
ADPGPP	GCTGGGTGGTAACGGGACTCGCCTCTTTCCTCTTACTAGCA	0023
Beta vulgaris	GGAGAGCTAAGCCAGCAGTGCCAATTGGAGG	
Ala98Lys	CCTCCAATTGGCACTGCTGGCTTAGCTCTCCTGCTAGTAAGAGGA	6030
GCT-AAC	AAGAGGCGAGTCCC <u>GTT</u> ACCACCACCCAGCACAATTGCAGCCAC	
	ATTTTTTGGGTCAGCTTTTGGAGATTCAAATA	
	TGGTGGT <u>AAC</u> GGGACTC	6031
	GAGTCCC <u>GTT</u> ACCACCA	6032
Increased Starch	CTAGCAGGAGAGCTAAGCCAGCAGTGCCAATTGGAGGGTGTTAC	6033
ADPGPP	AGGCTGATTGATGTGCTTATGAGCAACTGCATCAACAGTGGCATT	
Beta vulgaris Pro126Leu	AGAAAGATTTTCATTCTTACCCAGTTCAATTC	L

Pro126Leu CCT-CTT

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	GAATTGAACTGGGTAAGAATGAAAATCTTTCTAATGCCACTGTTGA TGCAGTTGCTCATAAGCACATCAATCAGCCTGTAACACCCTCCAA TTGGCACTGCTGGCTTAGCTCTCCTGCTAG	6034
	TGATGTGCTTATGAGCA	6035
	TGCTCATAAGCACATCA	6036
Increased Starch ADPGPP	CCCAGTTCAATTCGTTTTCGCTTAATCGTCATCTTGCTCGAACCTA TAATTTTGGAGATAATGTGAATTTTGGGGATGGCTTTGTGGAGGTT TTTGCTGCTACACACACACCCTGGAGAATC	6037
Beta vulgaris Gly162Asn GGT-AAT	GATTCTCCAGGTGTTTGTGTAGCAGCAAAAACCTCCACAAAGCCA TCCCCAAAATTCACATTATCTCCAAAATTATAGGTTCGAGCAAGAT GACGATTAAGCGAAAACGAATTGAACTGGG	6038
	TGGAGATAAGCGAAAACGATT TGAGAGATT	6039
	AATTCACATTATCTCCA	6040
Increased Starch ADPGPP	CCCAGTTCAATTCGTTTTCGCTTAATCGTCATCTTGCTCGAACCTA TAATTTTGGAGAT AAC GTGAATTTTGGGGGATGGCTTTGTGGAGGT	6041
Beta vulgaris Gly162Asn GGT-AAC	TTTTGCTGCTACACAAACACCTGGAGAATC GATTCTCCAGGTGTTTGTGTAGCAGCAAAAACCTCCACAAAGCCA TCCCCAAAATTCACCAAATTCAACTGGG CAAAATTCAACCAAATTCAACTGGG	6042
	GACGATTAAGCGAAACGAATTGAACTGGG TGGAGAT <u>AAC</u> GTGAATT	6043
	AATTCACGTTATCTCCA	6044

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Table 21
Oligonucleotides to produce plants with waxy starch

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS Arabidopsis thaliana	GAATCCAGGTAAACGGGTAGTTCATAATGGCAACTGTGACTGCTTC TTCTAACTTTGTGTGAAGAACTTCACTTTTCAACAATCATGGTGCTT CTTCATGCTCTGATGTCGCTCAGATTAC	6045
Ser12Term TCA-TGA	GTAATCTGAGCGACATCAGAGCATGAAGAAGCACCATGATTGTTGA AAAGTGAAGTTCTT <u>C</u> ACACAAAGTTAGAAGAAGCAGTCACAGTTGC CATTATGAACTACCCGTTTACCTGGATTC	6046
	CTTTGTGT <u>G</u> AAGAACTT	6047
	AAGTTCTT <u>C</u> ACACAAAG	6048
Waxy starch GBSS Arabidopsis thaliana	ATCCAGGTAAACGGGTAGTTCATAATGGCAACTGTGACTGCTTCTT CTAACTTTGTGTCATGAACTTCACTTTTCAACAATCATGGTGCTTCT TCATGCTCTGATGTCGCTCAGATTACCT	6049
Arg13Term AGA-TGA	AGGTAATCTGAGCGACATCAGAGCATGAAGAAGCACCATGATTGTT GAAAAGTGAAGTTCATGACACAAAGTTAGAAGAAGCAGTCACAGTT GCCATTATGAACTACCCGTTTACCTGGAT	6050
	TTGTGTCA <u>T</u> GAACTTCA	6051
	TGAAGTTC A TGACACAA	6052
Waxy starch GBSS Arabidopsis thaliana	TAAACGGGTAGTTCATAATGGCAACTGTGACTGCTTCTTCTAACTT TGTGTCAAGAACTTGACTTTTCAACAATCATGGTGCTTCTTCATGCT CTGATGTCGCTCAGATTACCTTAAAAGG	6053
Ser15Term TCA-TGA	CCTTTTAAGGTAATCTGAGCGACATCAGAGCATGAAGAAGCACCAT GATTGTTGAAAAGTCAAGTTCTTGACACAAAGTTAGAAGAAGCAGT CACAGTTGCCATTATGAACTACCCGTTTA	6054
	AAGAACTT G ACTTTTCA	6055
	TGAAAAGT <u>C</u> AAGTTCTT	6056
Waxy starch GBSS Arabidopsis thaliana	TGACTGCTTCTTCTAACTTTGTGTCAAGAACTTCACTTTTCAACAAT CATGGTGCTTCTTGATGTCGCTCAGATTACCTTAAAAG GCCAATCCTTGACTCATTGTGGGTTAAG	6057
Ser24Term TCA-TGA	CTTAACCCACAATGAGTCAAGGATTGGCCTTTTAAGGTAATCTGAG CGACATCAGAGCAT <u>C</u> AAGAAGCACCATGATTGTTGAAAAGTGAAGT TCTTGACACAAAGTTAGAAGAAGCAGTCA	6058
	TGCTTCTT <u>G</u> ATGCTCTG	6059
	CAGAGCATCAAGAAGCA	6060

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Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
Alteration /axy starch BSS	GTGCTTCTTCAACTTTGTGTCAAGAACTTCACTTTAAAAGGCCA	6061
rabidopsis thaliana sys25Term GC-TGA	ATCCTTGACTCATTGTGGGTTAAGGTCA TGACCTTAACCCACAATGAGTCAAGGATTGGCCTTTTAAGGTAATC TGAGCGACATCAGATCATGAAGAAGCACCATGATTGTTGAAAAAGTG AAGTTCTTGACACAAAGTTAGAAGAAGCA	6062
	TCTTCATGATCTGATGT	6063
	ACATCAGA <u>T</u> CATGAAGA	6064
Vaxy starch GBSS	GTAACAGCTTCACAGTTGGTGTCACATGTCCATGGTGGAGCAACG TCTTCACCGGATACTTAAACAAACTTGGCCCAGGTTGGCCTCAGG AACCAGCAATTCACTCACAATGGGTTGAGAT	6065
Antirrhinum majus ₋ ys24Term AAA-TAA	ACCAGCATTCACTOACTOAC	.6066
	CGGATACTTAAACAAAC	6067
	GTTTGTTT <u>A</u> AGTATCCG	6068
Waxy starch GBSS	CACAGTTGGTGTCACATGTCCATGGTGGAGCAACGTCTTCACCGG ATACTAAAACAAACTAGGCCCAGGTTGGCCTCAGGAACCAGCAAT TCACTCACAATGGGTTGAGATCAATAAACAT	6069
Antirrhinum majus Leu27Term TTG-TAG	ATGTTTATTGATCTCAACCCATTGTGAGTGAATTGCTGGTTCCTGA GGCCAACCTGGGCCTAGTTTGTTTTAGTATCCGGTGAAGACGTTG	6070
	CTCCACCATGGACATGTGACACCAACTGTG AACAAACTAGGCCCAGG	6071
	CCTGGGCCTAGTTTGTT	6072
Waxy starch GBSS	TTGGTGTCACATGTCCATGGTGGAGCAACGTCTTCACCGGATACT	6073
Antirrhinum majus Gln29Term CAG-TAG	CACAATGGGTTGAGATCAATAAACATGGTTG CAACCATGTTTATTGATCTCAACCCATTGTGAGTGAATTGCTGGTT CCTGAGGCCAACCTAGGCCAAGTTTGTTTTAGTATCCGGTGAAGA CGTTGCTCCACCATGGACATGTGACACCAA	607
	ACTTGGCC <u>T</u> AGGTTGGC	607
	GCCAACCT <u>A</u> GGCCAAGT	607
Waxy starch GBSS	GGTGGAGCAACGTCTTCACCGGATACTAAAACAAACTTGGCCCAG GTTGGCCTCAGGAACTAGCAATTCACTCACAATGGGTTGAGATCAA TAAACATGGTTGATAAGCTTCAAATGAGGA	1
Antirrhinum majus Gln35Term CAG-TAG	TCCTCATTTGAAGCTTATCAACCATGTTTATTGATCTCAACCCATTG TGAGTGAATTGCTAGTTCCTGAGGCCAACCTGGGCCAAGTTTGTT TTAGTATCCGGTGAAGACGTTGCTCCACC	607

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	TCAGGAAC <u>T</u> AGCAATTC	6079
	GAATTGCT <u>A</u> GTTCCTGA	6080
Waxy starch GBSS Antirrhinum majus	GGAGCAACGTCTTCACCGGATACTAAAACAAACTTGGCCCAGGTT GGCCTCAGGAACCAGTAATTCACTCACAATGGGTTGAGATCAATAA ACATGGTTGATAAGCTTCAAATGAGGAACA	6081
Gln36Term CAA-TAA	TGTTCCTCATTTGAAGCTTATCAACCATGTTTATTGATCTCAACCCA TTGTGAGTGAATTACTGGTTCCTGAGGCCAACCTGGGCCAAGTTT GTTTTAGTATCCGGTGAAGACGTTGCTCC	6082
	GGAACCAG <u>T</u> AATTCACT	6083
	AGTGAATT <u>A</u> CTGGTTCC	6084
Waxy starch GBSS Ipomoea batatas	GTGATGGCGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTG GGGGTGCCACTTCTTGAGAATCAAAAGTGGGGTTGGGTCAATTAG CCCTGAGGAGCCAAGCTGTGACTCACAATG	6085
Gly20Term GGA-TGA	CATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGACCCAACC CCACTTTTGATTCTCAAGAAGTGGCACCCCCACAGACATGAGAAA CAAAGTGTGAGGCAGTTATAGTCGCCATCAC	6086
	CCACTTCT <u>T</u> GAGAATCA	6087
	TGATTCTC A AGAAGTGG	6088
Waxy starch GBSS Ipomoea batatas	ATGGCGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGG GTGCCACTTCTGGATAATCAAAAGTGGGGTTGGGTCAATTAGCCC TGAGGAGCCAAGCTGTGACTCACAATGGGT	6089
Glu21Term GAA-TAA	ACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGACCCA ACCCCACTTTTGATTATCCAGAAGTGGCACCCCCACAGACATGAG AAACAAAGTGTGAGGCAGTTATAGTCGCCAT	6090
	CTTCTGGA <u>T</u> AATCAAAA	6091
	TTTTGATT <u>A</u> TCCAGAAG	6092
Waxy starch GBSS Ipomoea batatas	CGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGGGTGC CACTTCTGGAGAATGAAAAGTGGGGTTGGGTCAATTAGCCCTGAG GAGCCAAGCTGTGACTCACAATGGGTTGAG	6093
Ser22Term TCA-TGA	CTCAACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGA CCCAACCCCACTTTTCATTCTCCAGAAGTGGCACCCCCACAGACA TGAGAAACAAAGTGTGAGGCAGTTATAGTCG	6094
	TGGAGAAT <u>G</u> AAAAGTGG	6095
	CCACTITI C ATTCTCCA	6096

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS	ACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGGGTGCCA CTTCTGGAGAATCATAAGTGGGGTTGGGTCAATTAGCCCTGAGGA GCCAAGCTGTGACTCACAATGGGTTGAGAC	6097
oomoea batatas .ys23Term AAA-TAA	GTCTCAACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATT GACCCAACCCCACTTATGATTCTCCAGAAGTGGCACCCCCACAGA CATGAGAAACAAAGTGTGAGGCAGTTATAGT	6098
	GAGAATCA <u>T</u> AAGTGGGG	6099
	CCCCACTT <u>A</u> TGATTCTC	6100
Waxy starch	CCTCACACTTTGTTTCTCATGTCTGTGGGGGTGCCACTTCTGGAGA ATCAAAAGTGGGGTAGGGTCAATTAGCCCTGAGGAGCCAAGCTGT GACTCACAATGGGTTGAGACCTGTGAACAA	6101
Ipomoea batatas Leu26Term TTG-TAG	TTGTTCACAGGTCTCAACCCATTGTGAGTCACAGCTTGGCTCCTCA GGGCTAATTGACCCTACCCCACTTTTGATTCTCCAGAAGTGGCAC CCCCACAGACATGAGAAACAAAGTGTGAGG	6102
	AGTGGGTAGGGTCAAT	6103
	ATTGACCC <u>T</u> ACCCCACT	6104
Waxy starch GBSS	CATCGGCGATTGTTGCTCCTTACTGCTCTCTCACAGAATGGCAAC GGTGACGGGGTCTTAGGTGGTGTCGAGAAGCGCGTGCTTCAATTC CCAGGGAAGAACAGAAGCCAAAGTGAATTCA	6105
Astragalus membranaeus Tyr8Term	TGAATTCACTTTGGCTTCTGTTCTTCCCTGGGAATTGAAGCACGCG CTTCTCGACACCACCTAAGACCCCGTCACCGTTGCCATTCTGTGA GAGAGCAGTAAGGAGCAACAATCGCCGATG	6106
TAT-TAG	GGGTCTTAGGTGTC	6107
	GACACCAC <u>C</u> TAAGACCC	6108
Waxy starch GBSS Astragalus membranaeus Ser11Term	ATTGTTGCTCCTTACTGCTCTCTCACAGAATGGCAACGGTGACGG GGTCTTATGTGGTGTAGAAGAAGCGCGTGCTTCAATTCCCAGGGAA GAACAGAAGCCAAAGTGAATTCACCTCAGAA	6109
	TTCTGAGGTGAATTCACTTTGGCTTCTGTTCTTCCCTGGGAATTGA AGCACGCGCTTCTCTACACCACATAAGACCCCGTCACCGTTGCCA TTCTGTGAGAGAGCAGTAAGGAGCAACAAT	
TCG-TAG	TGTGGTGTAGAGAGCG	611
	CGCTTCTCTACACCACA	611

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	Phanotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Waxy starch GBSS Astragalus	TGTTGCTCCTTACTGCTCTCTCACAGAATGGCAACGGTGACGGGG TCTTATGTGGTGTCG <u>T</u> GAAGCGCGTGCTTCAATTCCCAGGGAAGA ACAGAAGCCAAAGTGAATTCACCTCAGAAGA	6113
5	membranaeus Arg12Term AGA-TGA	TCTTCTGAGGTGAATTCACTTTGGCTTCTGTTCTTCCCTGGGAATT GAAGCACGCGCTTCACGACACACATAAGACCCCGTCACCGTTGC CATTCTGTGAGAGAGCAGTAAGGAGCAACA	6114
		TGGTGTCG <u>T</u> GAAGCGCG	6115
	<u>.</u>	CGCGCTTC <u>A</u> CGACACCA	6116
	Waxy starch GBSS Astragalus	ACTGCTCTCACAGAATGGCAACGGTGACGGGGTCTTATGTGGT GTCGAGAAGCGCGTGATTCAATTCCCAGGGAAGAACAGAAGCCAA AGTGAATTCACCTCAGAAGATAAATCTCAAT	6117
10	membranaeus Cys15Term TGC-TGA	ATTGAGATTTATCTTCTGAGGTGAATTCACTTTGGCTTCTGTTCTTC CCTGGGAATTGAATCACGCGCTTCTCGACACCACATAAGACCCCG TCACCGTTGCCATTCTGTGAGAGAGCAGT	6118
		AGCGCGTG <u>A</u> TTCAATTC	6119
		GAATTGAA <u>T</u> CACGCGCT	6120
15	Waxy starch GBSS Astragalus	CACAGAATGGCAACGGTGACGGGGTCTTATGTGGTGTCGAGAAGC GCGTGCTTCAATTCC <u>T</u> AGGGAAGAACAGAAGCCAAAGTGAATTCA CCTCAGAAGATAAATCTCAATAGCCAAGCAT	6121
	membranaeus Gln19Term CAG-TAG	ATGCTTGGCTATTGAGATTTATCTTCTGAGGTGAATTCACTTTGGCT TCTGTTCTTCCCTAGGAATTGAAGCACGCGCTTCTCGACACCACAT AAGACCCCGTCACCGTTGCCATTCTGTG	6122
		TCAATTCC <u>T</u> AGGGAAGA	6123
		TCTTCCCT <u>A</u> GGAATTGA	6124
20	Waxy starch GBSS Solanum tuberosum	TGTAGCTTGGTAGATTCCCCTTTTTGTAGACCACACATCACATGGC AAGCATCACAGCTTGACACCACTTTGTGTCAAGAAGCCAAACTTCA CTAGACACCAAATCAACCTTGTCACAGAT	6125
	Ser7Term TCA-TGA	ATCTGTGACAAGGTTGATTTGGTGTCTAGTGAAGTTTGGCTTCTTG ACACAAAGTGGTGTCAAGCTGTGATGCTTGCCATGTGATGTGTGG TCTACAAAAAGGGGAATCTACCAAGCTACA	6126
		CACAGCTT <u>G</u> ACACCACT	6127
		AGTGGTGT <u>C</u> AAGCTGTG	6128
25	Waxy starch GBSS Solanum tuberosum	TCCCCTTTTGTAGACCACACATCACATGGCAAGCATCACAGCTTC ACACCACTTTGTGTGAAGAAGCCAAACTTCACTAGACACCAAATCA ACCTTGTCACAGATAGGACTCAGGAACCA	6129
	Ser12Term TCA-TGA	TGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGTGTCTAGTG AAGTTTGGCTTCTTCACACAAAGTGGTGTGAAGCTGTGATGCTTGC CATGTGATGTG	6130

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	CTTTGTGT G AAGAAGCC	6131
	GGCTTCTTCACACAAAG	6132
Vaxy starch GBSS	CCCTTTTGTAGACCACACATCACATGGCAAGCATCACAGCTTCAC ACCACTTTGTGTCATGAAGCCAAACTTCACTAGACACCAAATCAAC	6133
Solanum tuberosum Arg13Term AGA-TGA	CTTGTCACAGATAGGACTCAGGAACCATA TATGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGTGTCTAG TGAAGTTTGGCTTCATGACACAAAGTGGTGTGAAGCTGTGATGCTT GCCATGTGATGTG	6134
	TTGTGTCATGAAGCCAA	6135
	TTGGCTTCATGACACAA	6136
Waxy starch GBSS	TTGTAGACCACACATCACATGGCAAGCATCACAGCTTCACACCACT TTGTGTCAAGAAGCTAAACTTCACTAGACACCAAATCAACCTTGTC	6137
Solanum tuberosum Gln15Term CAA-TAA	ACAGATAGGACTCAGGAACCATACTCTGA TCAGAGTATGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGT GTCTAGTGAAGTTTAGCTTCTTGACACAAAGTGGTGTGAAGCTGTG	6138
	ATGCTTGCCATGTGATGTGTGGTCTACAA CAAGAAGC <u>T</u> AAACTTCA	6139
	TGAAGTITAGCTTCTTG	6140
Waxy starch GBSS	CCACACATCACATGGCAAGCATCACAGCTTCACACCACTTTGTGTC AAGAAGCCAAACTTGACTAGACACCAAATCAACCTTGTCACAGATA GGACTCAGGAACCATACTCTGACTCACAA	614′
Solanum tuberosum Ser17Term TCA-TGA	TTGTGAGTCAGAGCCATACTCTGACTCAGTCACTCTGTGACAAGGTTG ATTTGGTGTCTAGTCAAGTTTGGCTTCTTGACACAAAGTGGTGTGA AGCTGTGATGCTTGCCATGTGATGTG	6142
	CCAAACTT <u>G</u> ACTAGACA	614
	TGTCTAGT <u>C</u> AAGTTTGG	614
Waxy starch GBSS	GTCGATCACTCTTCTCTCACCGCCGAAACAGATTTTGACACAAAAA TGGCAACAATAACGTGATCTTCAATGCCGACGAGAACCGCGTGCT TCAATTACCAAGGAAGATCAGCAGAGTCTA	614
Pisum sativum Gly6Term GGA-TGA	TAGACTCTGCTGATCTTCCTTGGTAATTGAAGCACGCGGTTCTCGT CGGCATTGAAGATCACGTTATTGTTGCCATTTTTGTGTCAAAATCT	614
	GTTTCGGCGGTGAGAGAGAGTGATCGAC CAATAACGTGATCTTCA	614
	TGAAGATCACGTTATTG	614

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS Pisum sativum	ACTCTTCTCACCGCCGAAACAGATTTTGACACAAAAATGGCAAC AATAACGGGATCTTGAATGCCGACGAGAACCGCGTGCTTCAATTA CCAAGGAAGATCAGCAGAGTCTAAACTGAA	6149
Ser8Term TCA-TGA	TTCAGTTTAGACTCTGCTGATCTTCCTTGGTAATTGAAGCACGCGG TTCTCGTCGGCATTCAAGATCCCGTTATTGTTGCCATTTTTGTGTC AAAATCTGTTTCGGCGGTGAGAGAGAGAGT	6150
	GGGATCTT <u>G</u> AATGCCGA	6151
	TCGGCATT <u>C</u> AAGATCCC	6152
Waxy starch GBSS Pisum sativum	ACCGCCGAAACAGATTTTGACACAAAAATGGCAACAATAACGGGAT CTTCAATGCCGACGTGAACCGCGTGCTTCAATTACCAAGGAAGAT CAGCAGAGTCTAAACTGAATTTGCCTCAGA	6153
Arg12Term AGA-TGA	TCTGAGGCAAATTCAGTTTAGACTCTGCTGATCTTCCTTGGTAATT GAAGCACGCGGTTCACGTCGGCATTGAAGATCCCGTTATTGTTGC CATTTTTGTGTCAAAATCTGTTTCGGCGGT	6154
	TGCCGACG <u>T</u> GAACCGCG	6155
	CGCGGTTC <u>A</u> CGTCGGCA	6156
Waxy starch GBSS <i>Pisum sativum</i>	AGATTTTGACACAAAAATGGCAACAATAACGGGATCTTCAATGCCG ACGAGAACCGCGTGATTCAATTACCAAGGAAGATCAGCAGAGTCT AAACTGAATTTGCCTCAGATACACTTCAAT	6157
Cys15Term TGC-TGA	ATTGAAGTGTATCTGAGGCAAATTCAGTTTAGACTCTGCTGATCTT CCTTGGTAATTGAATCACGCGGTTCTCGTCGGCATTGAAGATCCC GTTATTGTTGCCATTTTTGTGTCAAAATCT	6158
	ACCGCGTG <u>A</u> TTCAATTA	6159
	TAATTGAA <u>T</u> CACGCGGT	6160
Waxy starch GBSS Pisum sativum	CACAAAAATGGCAACAATAACGGGATCTTCAATGCCGACGAGAAC CGCGTGCTTCAATTAGCAAGGAAGATCAGCAGAGTCTAAACTGAAT TTGCCTCAGATACACTTCAATAACAACCAA	6161
Tyr18Term TAC-TAG	TTGGTTGTTATTGAAGTGTATCTGAGGCAAATTCAGTTTAGACTCTG CTGATCTTCCTTGCTAATTGAAGCACGCGGTTCTCGTCGGCATTGA AGATCCCGTTATTGTTGCCATTTTTGTG	6162
	TTCAATTA <u>G</u> CAAGGAAG	6163
	CTTCCTTG <u>C</u> TAATTGAA	6164
Waxy starch GBSS Manihot esculenta	TCTACACCGGAGAGAGCACCATGGCAACTGTAATAGCTGCACATT TCGTTTCCAGGAGCTGACACTTGAGCATCCATGCATTAGAGACTAA GGCTAATAATTTGTCTCACACTGGACCCTG	6165
Ser14Term TCA-TGA	CAGGGTCCAGTGTGAGACAAATTATTAGCCTTAGTCTCTAATGCAT GGATGCTCAAGTGTCAGCTCCTGGAAACGAAATGTGCAGCTATTA CAGTTGCCATGGTGCTCTCCCGGTGTAGA	6166

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	CAGGAGCT G ACACTTGA	6167
	TCAAGTGTCAGCTCCTG	6168
Vaxy starch GBSS	CCGGAGAGCACCATGGCAACTGTAATAGCTGCACATTTCGTTT CCAGGAGCTCACACTAGAGCATCCATGCATTAGAGACTAAGGCTA	6169
Manihot esculenta Leu16Term ITG-TAG	ATAATTTGTCTCACACTGGACCCTGGACCCA TGGGTCCAGGGTCCAGTGTGAGACAAATTATTAGCCTTAGTCTCTA ATGCATGGATGCTCTAGTGTGAGACCAAACGAAATGTGCAG	6170
	CTATTACAGTTGCCATGGTGCTCTCCCGG CTCACACTAGAGCATCC	6171
	GGATGCTC <u>T</u> AGTGTGAG	6172
Waxy starch GBSS	TGGCAACTGTAATAGCTGCACATTTCGTTTCCAGGAGCTCACACTT GAGCATCCATGCATGAGAGACTAAGGCTAATAATTTGTCTCACACT GGACCCTGGACCCAAACTATCACTCCCAA	6173
<i>Manihot esculenta</i> Leu21Term TTA-TGA	TTGGACCCTAACTATCACTCCCAX TTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAGACAAATTA TTAGCCTTAGTCTCTCATGCATGGATGCTCAAGTGTGAGCTCCTGG AAACGAAATGTGCAGCTATTACAGTTGCCA	6174
	CCATGCATGAGAGACTA	6175
	TAGTCTCTCATGCATGG	6176
Waxy starch GBSS	GCAACTGTAATAGCTGCACATTTCGTTTCCAGGAGCTCACACTTGA GCATCCATGCATTATAGACTAAGGCTAATAATTTGTCTCACACTGG ACCCTGGACCCAAACTATCACTCCCAATG	6177
Manihot esculenta Glu22Term GAG-TAG	CATTGGGACCCAAACTATCACTCGCATTG CATTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAGACAAAT TATTAGCCTTAGTCTATAATGCATGGATGCTCAAGTGTGAGCTCCT GGAAACGAAATGTGCAGCTATTACAGTTGC	6178
	ATGCATTA <u>T</u> AGACTAAG	6179
	CTTAGTCT <u>A</u> TAATGCAT	6180
Waxy starch GBSS	GTAATAGCTGCACATTTCGTTTCCAGGAGCTCACACTTGAGCATCC ATGCATTAGAGACTTAGGCTAATAATTTGTCTCACACTGGACCCTG GACCCAAACTATCACTCCCAATGGTTTAA	618
Manihot esculenta Lys24Term AAG-TAG	TTAAACCATTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAG ACAAATTATTAGCCTAAGTCTCTAATGCATGGATGCTCAAGTGTGA	618
	GCTCCTGGAAACGAAATGTGCAGCTATTAC TAGAGACTTAGGCTAAT	618
	ATTAGCCTAAGTCTCTA	618

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS Phaseolus vulgaris	ACAACTCCTCCGTCACCGGTATAAGCATGGCAACGGTATCGATGG CATCGTGCGTGGCGTG	6185
Ser12Term TCA-TGA	CGGTTCAGGCTCATCTGACCCGAAGATTTCACTTTTGTCTCTGTAC TCCACGCGCCTTTTCACGCCACGC	6186
	CGTGGCGT <u>G</u> AAAAGGCG	6187
	CGCCTTTT <u>C</u> ACGCCACG	6188
Waxy starch GBSS Phaseolus vulgaris	CACCGGTATAAGCATGGCAACGGTATCGATGGCATCGTGCGTG	6189
Trp16Term TGG-TGA	TTTCAATTCATGACGGTTCAGGCTCATCTGACCCGAAGATTTCACT TTTGTCTCTGTACT <u>T</u> CACGCGCCTTTTGACGCCCACGCACGATGCC ATCGATACCGTTGCCATGCTTATACCGGTG	6190
	GGCGCGTG <u>A</u> AGTACAGA	6191
	TCTGTACT <u>T</u> CACGCGCC	6192
Waxy starch GBSS Phaseolus vulgaris	ATAAGCATGGCAACGGTATCGATGGCATCGTGCGTGGCGTCAAAA GGCGCGTGGAGTACA <u>T</u> AGACAAAAGTGAAATCTTCGGGTCAGATG AGCCTGAACCGTCATGAATTGAAATACGATG	6193
Glu19Term GAG-TAG	CATCGTATTTCAATTCATGACGGTTCAGGCTCATCTGACCCGAAGA TTTCACTTTTGTCTATGTACTCCACGCGCCTTTTGACGCCACGCAC GATGCCATCGATACCGTTGCCATGCTTAT	6194
	GGAGTACA <u>T</u> AGACAAAA	6195
	TTTTGTCT <u>A</u> TGTACTCC	6196
Waxy starch GBSS Phaseolus vulgaris	ATGGCAACGGTATCGATGGCATCGTGCGTGGCGTCAAAAGGCGC GTGGAGTACAGAGACATAAGTGAAATCTTCGGGTCAGATGAGCCT GAACCGTCATGAATTGAAATACGATGGGTTGA	6197
Lys21Term AAA-TAA	TCAACCCATCGTATTTCAATTCATGACGGTTCAGGCTCATCTGACC CGAAGATTTCACTTATGTCTCTGTACTCCACGCGCCTTTTGACGCC ACGCACGATGCCATCGATACCGTTGCCAT	6198
	CAGAGACA <u>T</u> AAGTGAAA	6199
	TTTCACTT <u>A</u> TGTCTCTG	6200
Waxy starch GBSS Phaseolus vulgaris	ACGGTATCGATGGCATCGTGCGTGGCGTCAAAAGGCGCGTGGAG TACAGAGACAAAAGTGTAATCTTCGGGTCAGATGAGCCTGAACCG TCATGAATTGAAATACGATGGGTTGAGATCTC	6201
Lys23Term AAA-TAA	GAGATCTCAACCCATCGTATTTCAATTCATGACGGTTCAGGCTCAT CTGACCCGAAGATTACACTTTTGTCTCTGTACTCCACGCGCCTTTT GACGCCACGCACGATGCCATCGATACCGT	6202

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	CAAAAGTG <u>T</u> AATCTTCG	6203
	CGAAGATT <u>A</u> CACTTTTG	6204
Vaxy starch GBSS	GCGCCTAGCTCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGG GTTCCATTCCTAATTAGTGTTCTTATCAAACAACAGTGTTGGTTCA	6205
Г <i>riticum aestivum</i> Гуг7Тегт ГАТ-ТАG	CTGAAACTGTCGCCTCACATCCAATTCCAG CTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACCAACACTGTTT GTTTGATAAGAACACTAATTAGGAATGGAACCCATTGGTGCAGCCT CTCAATGACGACCTTTTCGAGCTAGGCGC	6206
	CCTAATTAGTGTTCTTA	6207
	TAAGAACA <u>C</u> TAATTAGG	6208
Waxy starch GBSS	CCTAGCTCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTC CATTCCTAATTATTGATCTTATCAAACAACAGTGTTGGTTCACTGA	6209
<i>Triticum aestivum</i> Cys8Term TGT-TGA	AACTGTCGCCTCACATCCAATTCCAGCAA TTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACCAACACT GTTTGTTTGATAAGATCAATAATTAGGAATGGAACCCATTGGTGCA	6210
	GCCTCTCAATGACGACCTTTTCGAGCTAGG AATTATTGATCTTATCA	6211
•	TGATAAGA <u>T</u> CAATAATT	6212
Waxy starch GBSS	TCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTCCATTCC TAATTATTGTTCTTAGCAAACAAACAGTGTTGGTTCACTGAAACTGT	6213
Triticum aestivum Tyr10Term TAT-TAG	CGCCTCACATCCAATTCCAGCAATCTTGT ACAAGATTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACC AACACTGTTTGTTTGCTAAGAACAATAATTAGGAATGGAACCCATT GGTGCAGCCTCTCAATGACGACCTTTTCGA	6214
	TGTTCTTAGCAAACAAA	621
	TTTGTTTGCTAAGAACA	621
Waxy starch GBSS	CGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTCCATTCCT AATTATTGTTCTTATTAAACAAACAGTGTTGGTTCACTGAAACTGTC GCCTCACATCCAATTCCAGCAATCTTGTA	621
Triticum aestivum Gln11Term CAA-TAA	TACAAGATTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAAC CAACACTGTTTGTTTAATAAGAACAATAATTAGGAATGGAACCCATT	621
	GGTGCAGCCTCTCAATGACGACCTTTTCG GTTCTTATTAAACAAAC	621
	GTTTGTTT A ATAAGAAC	622

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS Triticum aestivum	AGGCTGCACCAATGGGTTCCATTCCTAATTATTGTTCTTATCAAACA AACAGTGTTGGTTGACTGAAACTGTCGCCTCACATCCAATTCCAGC AATCTTGTAACAATGAAGTTATGTTCCT	6221
Ser17Term TCA-TGA	AGGAACATAACTTCATTGTTACAAGATTGCTGGAATTGGATGTGAG GCGACAGTTTCAGTCAACCAACACTGTTTGTTTGATAAGAACAATA ATTAGGAATGGAACCCATTGGTGCAGCCT	6222
	TGTTGGTT G ACTGAAAC	6223
	GTTTCAGT C AACCAACA	6224
Waxy starch GBSS Triticum aestivum	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTGCAGGTTTCTAGGGCGTGAGGCCCCGGAGCCCGGCG GATGCGGCTCTCGGCATGAGGACCGTCGGAGCTA	6225
Gln28Term CAG-TAG	TAGCTCCGACGGTCCTCATGCCGAGAGCCGCATCCGCCGGGCTCCGGGGCCTCACGCCCTAGAAACCTGCACGCCGGAACCTGTCGGTGATGCCGAGGACCGGTGCCGGAGGTGGCGAGCTG	6226
	CAGGTTTC <u>T</u> AGGGCGTG	6227
	CACGCCCT <u>A</u> GAAACCTG	6228
Waxy starch GBSS Triticum aestivum	GGTTTCCAGGGCGTGAGGCCCGGAGCCCGGCGGATGCGGCTC TCGGCATGAGGACCGTCTGAGCTAGCGCCGCCCCAACGCAAAGC CGGAAAGCGCACCGCGGGACCCGGCGGTGCCTCT	6229
Gly46Term GGA-TGA	AGAGGCACCGCGGGTCCCGCGGTGCGCTTTCCGGCTTTGCGTT GGGGCGCGCTAGCTCAGACCGGTCCTCATGCCGAGAGCCGCATC CGCCGGGCTCCGGGGCCTCACGCCCTGGAAACC	6230
	GGACCGTC <u>T</u> GAGCTAGC	6231
	GCTAGCTC <u>A</u> GACGGTCC	6232
Waxy starch GBSS Triticum aestivum	CGGAGCCCGGCGATGCGGCTCTCGGCATGAGGACCGTCGGAG CTAGCGCCGCCCCAACGTAAAGCCGGAAAGCGCACCGCGGGACC CGGCGGTGCCTCTCCATGGTGGTGCGCGCCACCG	6233
Gin53Term CAA-TAA	CGGTGGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTACGTTGGGGCGCGCTAGCTCCGACGG TCCTCATGCCGAGAGCCGCATCCGCCGGGCTCCG	6234
	CCCCAACGTAAAGCCGG	6235
	CCGGCTTTACGTTGGGG	6236
Waxy starch GBSS <i>Triticum aestivum</i>	GCGGATGCGGCTCTCGGCATGAGGACCGTCGGAGCTAGCGCCG CCCCAACGCAAAGCCGGTAAGCGCACCGCGGGACCCGGCGGTG CCTCTCCATGGTGGTGCGCGCCACCGGCAGCGGCG	6237
Lys56Term AAA-TAA	CGCCGCTGCCGGTGGCGCGCACCACCATGGAGAGGCACCGCCG GGTCCCGCGGTGCGCTTACCGGCTTTGCGTTGGGGCGCGCTAG CTCCGACGGTCCTCATGCCGAGAGCCGCATCCGC	6238

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	AAAGCCGG <u>T</u> AAGCGCAC	6239
	GTGCGCTTACCGGCTTT	6240
Vaxy starch	CTCTCCATGGTGGTGCGCGCCACCGGCAGCGGCGCATGAACCT CGTGTTCGTCGGCGCCTAGATGGCGCCCTGGAGCAAGACCGGCG	6241
BBSS Friticum aestivum Bu85Term BAG-TAG	CTCGGCGACGTCCTCGGGGGCCTCCCCCAG CTGGGGGGAGGCCCCCGAGGACGCCGAGGCCCGGTCTT GCTCCAGGGCGCCATCTAGGCGCCGACGAACACGAGGTTCATGC	6242
	CGCCGCTGCCGGTGGCGCACCACCATGGAGAG TCGGCGCCTAGATGGCG	6243
	CGCCATCTAGGCGCCGA	6244
Waxy starch GBSS	GTCGTCTCGCTGCAGGTAGCCACACCCTGCGCGCGCGATGGC GGCTCTGGTCACGTCGTAGCTCGCCACCTCCGGCACCGTCCTCG	6245
Triticum aestivum Gln8Term CAG-TAG	GCATCACCGACAGGTTCCGGCGTGCAGGTTTTC GAAAACCTGCACGCCGGAACCTGTCGGTGATGCCGAGGACGGTG CCGGAGGTGGCGAGCTACGACGTGACCAGAGCCGCCATCGCGC GCGCAGGGTGTGGCTACCTGCAGCGAGAGACGAC	6246
	TCACGTCGTAGCTCGCC	6247
	GGCGAGCT <u>A</u> CGACGTGA	6248
Waxy starch GBSS	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTGCAGGTTTTTAGGGTGTGAGGCCCCGGAGCCCGGCAG ATGCGCCGCTCGGCATGAGGACTACCGGAGCGA	6249
Triticum aestivum Gln28Term CAG-TAG	TCGCTCCGCATGAGGACTACCCCATCCCATCCCATCCCA	6250
	CAGGTTTT <u>T</u> AGGGTGTG	625
	CACACCCT <u>A</u> AAAACCTG	625
Waxy starch GBSS	CCCCGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCG GAGCGAGCGCCCCCGTAGCAACAAAGCCGGAAAGCGCACCG CGGGACCCGGCGGTGCCTCTCCATGGTGGTGCGCG	625
Triticum aestivum Lys52Term AAG-TAG	CGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTGCGC TTTCCGGCTTTGTTGCTACGGGGCGCGCGCTCGCTCCGGTAGTCC	625
	TCATGCCGAGCGCGCATCTGCCGGGCTCCGGGG CCGCCCGTAGCAACAA	625
	TTGTTGCTACGGGGCGG	625

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Waxy starch GBSS Triticum aestivum	CGGAGCCCGCAGATGCGCCGCTCGGCATGAGGACTACCGGAG CGAGCGCCCCCGAAG <u>T</u> AACAAAGCCGGAAAGCGCACCGCGG GACCCGGCGTGCCTCTCCATGGTGCTGCGCGCA	6257
GIn53Term CAA-TAA	TGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGCGCGCTCCGGTAG TCCTCATGCCGAGCGGCGCATCTGCCGGGCTCCG	6258
	CCCCGAAGTAACAAAGC	6259
	GCTTTGTT <u>A</u> CTTCGGGG	6260
Waxy starch GBSS Triticum aestivum	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAGCGA GCGCCGCCCCGAAGCAATAAAGCCGGAAAGCGCACCGCGGGAC CCGGCGTGCCTCTCCATGGTGCTGCGCGCCACGG	6261
Gln54Term CAA-TAA	CCGTGGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGCGCTCGCTC	6262
	CGAAGCAATAAAGCCGG	6263
	CCGGCTTTATTGCTTCG	6264
Waxy starch GBSS Triticum durum	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTGCAGGTTTCTAGGGCGTGAGGCCCCGGAACCCGGCG GATGCGGCCCTCGTCATGAGGACTATCGGAGCGA	6265
Gln28Term CAG-TAG	TCGCTCCGATAGTCCTCATGACGAGGGCCGCATCCGCCGGGTTC CGGGGCCTCACGCCCTAGAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6266
	CAGGTTTC <u>T</u> AGGGCGTG	6267
	CACGCCCT <u>A</u> GAAACCTG	6268
Waxy starch GBSS Triticum durum	CCCCGGAACCCGGCGGATGCGGCCCTCGTCATGAGGACTATCGG AGCGAGCGCCCCCGTAGCAAAGCCGGAAAGCGCACCGCGGG AGCCGGCGGTGCCTCTCCATGGTGGTGCGCGCCA	6269
Lys52Term AAG-TAG	TGGCGCGCACCATGGAGAGGCACCGCCGGCTCCCGCGGTG CGCTTTCCGGCTTTGCTACGGGGCGCGCGCTCCCGATAGTCC TCATGACGAGGCCGCATCCGCCGGGTTCCGGGG	6270
	CCGCCCGTAGCAAAGC	6271
	GCTTTGCT <u>A</u> CGGGGCGG	6272
Waxy starch GBSS Triticum durum	CGGAACCCGGCGGATGCGGCCCTCGTCATGAGGACTATCGGAGC GAGCGCCCCCGAAG <u>T</u> AAAGCCGGAAAGCGCACCGCGGGAGC CGGCGGTGCCTCTCCATGGTGGTGCGCGCCACGG	6273
Gln53Term CAA-TAA	CCGTGGCGCACCACCATGGAGAGGCACCGCCGGCTCCCGCG GTGCGCTTTCCGGCTTTACTTCGGGGCGCGCTCCGATAG TCCTCATGACGAGGGCCGCATCCGCCGGGTTCCG	6274

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	CCCCGAAG <u>T</u> AAAGCCGG	6275
	CCGCTTTACTTCGGGG	6276
Vaxy starch GBSS	GCGGATGCGGCCCTCGTCATGAGGACTATCGGAGCGAGCG	6277
riticum durum ys56Term AA-TAA	CTCTCCATGGTGCGCGCCACGGGCAGCGGCGCGCGCGCGC	6278
•	AAAGCCGGTAAGCGCAC	6279
	GTGCGCTTACCGGCTTT	6280
Waxy starch	TATCGGAGCGAGCGCCCCGAAGCAAAGCCGGAAAGCGCACC GCGGGAGCCGGCGGTGACTCTCCATGGTGGTGCGCGCCACGGG CAGCGGCGCATGAACCTCGTGTTCGTCGGCGCC	6281
<i>Triticum durum</i> Cys64Term TGC-TGA	GGCGCCGACGACACGAGGTTCATGCCGCCGCTGCCCGTGGCG	6282
	CCGCTTTGCTTCGGGGCGCGCTCGCTCCGATA CGGCGGTGACTCTCCAT	6283
	ATGGAGAG <u>T</u> CACCGCCG	6284
Waxy starch GBSS	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTGCAGGTTTTTAGGGTGTGAGGCCCCGGAGCCCGGCAG	6285
Triticum turgidum Gln28Term CAG-TAG	ATGCGCCGCTCGGCATGAGGACTACCGGAGCGA TCGCTCCGGTAGTCCTCATGCCGAGCGGCGCATCTGCCGGGCTC CGGGGCCTCACACCCTAAAAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGTGCCGGAGGTGGCGAGCTG	6286
	CAGGTTTT <u>T</u> AGGGTGTG	628
	CACACCCT <u>A</u> AAAACCTG	628
Waxy starch GBSS	CCCCGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCG GAGCGAGCGCCCCCGTAGCAACAAAGCCGGAAAGCGCACCG CGGGACCCGGCGGTGCCTCTCCATGGTGGTGCGCG	628
Triticum turgidum Lys52Term AAG-TAG	CGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTGCGCTTTCCCGGCTTTGTTGCTACGGGGCGCGCGC	629
	TCATGCCGAGCGCGCATCTGCCGGGGCTCCGGGG CCGCCCGTAGCAACAA	629
	TTGTTGCTACGGGGCGG	629

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS <i>Triticum turgidum</i>	CGGAGCCCGCAGATGCGCCGCTCGGCATGAGGACTACCGGAG CGAGCGCCGCCCGAAGTAACAAAGCCGGAAAGCGCACCGCGG GACCCGGCGGTGCCTCCATGGTGGTGCGCGCCA	6293
GIn53Term CAA-TAA	TGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGCGCCTCCGCTAG TCCTCATGCCGAGCGCGCATCTGCCGGGCTCCG	6294
	CCCCGAAG <u>T</u> AACAAAGC	6295
	GCTTTGTT <u>A</u> CTTCGGGG	6296
Waxy starch GBSS Triticum turgidum	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAGCGA GCGCCGCCCGAAGCAATAAAGCCGGAAAGCGCACCGCGGGAC CCGGCGGTGCCTCCATGGTGGTGCGCGCCACGG	6297
GIn54Term CAA-TAA	CCGTGGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGCGCTCCGCT TAGTCCTCATGCCGAGCGCGCGCATCTGCCGGGCT	6298
	CGAAGCAA <u>T</u> AAAGCCGG	6299
	CCGGCTTTATTGCTTCG	6300
Waxy starch GBSS Triticum turgidum	GATGCGCCGCTCGGCATGAGGACTACCGGAGCGAGCGCCCCCCCGAAGCAACAAAGCCGGTAAGCGCACCGCGGGACCCGGCGGTGCCCCCCATGGTGGTGCGCGCCACGGCCAGCGCCCG	6301
Lys57Term AAA-TAA	CGGCGCTGCCGTGGCGCGCACCACCATGGAGAGGCACCGCCG GGTCCCGCGGTGCGCTTACCGGCTTTGTTGCTTCGGGGCGCGC TCGCTCCGGTAGTCCTCATGCCGAGCGCGCATC	6302
	AAAGCCGG <u>T</u> AAGCGCAC	6303
	GTGCGCTT <u>A</u> CCGGCTTT	6304
Waxy starch GBSS Aegilops speltoides	CAGCTCGCCACCTCCGCCACCGTCCTCGGCATCACCGACAGGTT CCGCCATGCAGGTTTCTAGGGCGTGAGGCCCCGGAGCCCGGCAG ATGCGCCGCTCGGCATGAGGACTGTCGGAGCGA	6305
GIn28Term CAG-TAG	TCGCTCCGACAGTCCTCATGCCGAGCGCGCATCTGCCGGGCTC CGGGGCCTCACGCCCTAGAAACCTGCATGGCGAAACCTGTCGGT GATGCCGAGGACGGTGGCGAGGTGGCGAGCTG	6306
	CAGGTTTC <u>T</u> AGGGCGTG	6307
	CACGCCCT <u>A</u> GAAACCTG	6308
Waxy starch GBSS Aegilops speltoides	GGTTTCCAGGGCGTGAGGCCCCGGAGCCCGGCAGATGCGCCGC TCGGCATGAGGACTGTCTGAGCGAGCGCCCCCCGAAGCAACAA AGCCGGAAAGCGCACCGCGGGACCCGGCGGTGCC	6309
Gly46Term GGA-TGA	GGCACCGCCGGGTCCCGCGGTGCGCTTTCCGGCTTTGTTGCTTC GGGGCGCGCGC	6310

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	TGGACTGTCTGAGCGAGC	6311
	GCTCGCTCAGACAGTCC	6312
Vaxy starch	CCCCGGAGCCCGCAGATGCGCCGCTCGGCATGAGGACTGTCG GAGCGAGCGCCCCCGTAGCAACAAAGCCGGAAAGCGCACCG	6313
Aegilops speltoides Lys52Term AAG-TAG	CGGGACCCGGCGTGCCTCTCGATGGTGGTGCGCG CGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCGGTGCGC TTTCCGGCTTTGTTGCTACGGGGCGCGCTCCGGGG	6314
*	TCATGCCGAGCGCGCATCTGCCGGGCTCCGGGG CCGCCCCGTAGCAACAA	6315
	TTGTTGCTACGGGGCGG	6316
Waxy starch	CGAGCCCGCAGATGCGCCGCTCGGCATGAGGACTGTCGGAG	6317
Aegilops speltoides Gln53Term CAA-TAA	GACCCGGCGTGCCTCTCGATGGTGCGCGCCA TGGCGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGCGCTCCGACAG TCCTCATGCCGAGCGCGCATCTGCCGGGCTCCG	6318
	CCCGAAGTAACAAAGC	6319
	GCTTTGTT <u>A</u> CTTCGGGG	6320
Waxy starch GBSS	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTGTCGGAGCGA GCGCCGCCCGAAGCAATAAAGCCGGAAAGCGCACCGCGGGAC	6321
Aegilops speltoides GIn54Term CAA-TAA	CCGCCGTGCCTCTCGATGGTGGTGCGCGCCACCG CGGTGGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGCGCCTCCCGA CAGTCCTCATGCCGAGCGCGCATCTGCCGGGCT	6322
	CGAAGCAATAAAGCCGG	6323
	CCGGCTTTATTGCTTCG	6324
Waxy starch GBSS	AGTGCAGAGATCTTCCACAGCAACAGCTAGACAACCACCATGTCG GCTCTCACCACGTCCTAGCTCGCCACCTCGCCACCACGCTCGCCGCCGTCGTCGCCGCCGCCGCCGCCGCCGCCGC	632
Oryza glaberrima Gln8Term CAG-TAG	GCAGCGACGCGCGCCGACCTGTCAGCGATGCCGAAGCCGGT GGCCGAGGTGGCGAGCTAGGACGTGGTGAGAGCCGACATGGTG GTTGTCTAGCTGTTGCTGTGGAAGATCTCTGCACT	632
	CCACGTCCTAGCTCGCCC	632
	GGCGAGCTAGGACGTGG	632

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Phenotype, Gene, Plant & Targeted Afteration	Altering Oligos	SEQID NO:
Waxy starch GBSS Oryza glaberrima	TCCACAGCAACAGCTAGACAACCACCATGTCGGCTCTCACCACGT CCCAGCTCGCCACCTAGGCCACCGGCTTCGGCATCGCTGACAGG TCGGCGCCGTCGTCGCTGCTCCGCCACGGGTT	6329
Ser12Term TCG-TAG	AACCCGTGGCGAGCAGCGACGACGCGCCGACCTGTCAGCGAT GCCGAAGCCGGTGGCCTAGGTGGCGAGCTGGGACGTGAGA GCCGACATGGTGGTTGTCTAGCTGTTGCTGTGGA	6330
	CGCCACCT <u>A</u> GGCCACCG	6331
	CGGTGGCC <u>T</u> AGGTGGCG	6332
Waxy starch GBSS Oryza glaberrima	CGGCTCTCACCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTC GGCATCGCTGACAGGTAGGCCCCGCGCCGCCGCCGCCGCCGCCGCCGCCGCCGCCG	6333
Ser22Term TCG-TAG	CCGCCGGGCTGCGGGCTTGAGGCCCTGGAACCCGTGGCGA GCAGCGACGCGCCTACCTGTCAGCGATGCCGAAGCCGGTG GCCGAGGTGCCGAGCTGGGACGTGAGAGCCG	6334
	TGACAGGT <u>A</u> GGCGCCGT	6335
	ACGGCGCC <u>T</u> ACCTGTCA	6336
Waxy starch GBSS Oryza glaberrima	CCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCT GACAGGTCGCGCGCGTAGTCGCTGCTCCGCCACGGGTTCCAGG GCCTCAAGCCCCGCAGCCCCGCCGGCGCGACGC	6337
Ser25Term TCG-TAG	GCGTCGCCGGCGGGGCTGCGGGGCTTGAGGCCCTGGAACC CGTGGCGGAGCAGCGACTACGGCGCCGACCTGTCAGCGATGCC GAAGCCGGTGGCCGAGGTGGCGAGCTGGGACGTGG	6338
	GGCGCCGT <u>A</u> GTCGCTGC	6339
	GCAGCGAC <u>T</u> ACGGCGCC	6340
Waxy starch GBSS Oryza glaberrima	CGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCTGAC AGGTCGGCGCCGTCGTAGCTGCTCCGCCACGGGTTCCAGGGCCT CAAGCCCCGCAGCCCCGCCGGCGGCGACGCGAC	6341
Ser26Term TCG-TAG	GTCGCGTCGCCGGCGGGGCTGCGGGGCTTGAGGCCCTGGA ACCCGTGGCGAGCAGCTACGACGGCGCCGACCTGTCAGCGATG CCGAAGCCGGTGGCCGAGGTGGCGAGCTGGGACG	6342
	GCCGTCGT <u>A</u> GCTGCTCC	6343
	GGAGCAGC <u>T</u> ACGACGGC	6344
Waxy starch GBSS Oryza sativa	TCCACAGCAAGAGCTAAACAGCCGACCGTGTGCACCACCATGTCG GCTCTCACCACGTCCTAGCTCGCCACCTCGGCCACCGGCTTCGG CATCGCCGACAGGTCGGCGCCCGTCGTCGCTGC	6345
Gln8Term CAG-TAG	GCAGCGACGCGCCCGACCTGTCGGCGATGCCGAAGCCGGT GGCCGAGGTGGCGAGCTAGGACGTGGTGAGAGCCGACATGGTG GTGCACACGGTCGGCTGTTTAGCTCTTGCTGTGGA	6346

Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
Alteration	CCACGTCCTAGCTCGCC	6347
		6348
Vaxy starch	CTAAACAGCCGACCGTGTGCACCACCATGTCGGCTCTCACCACGT CCCAGCTCGCCACCTAGGCCACCGGCTTCGGCATCGCCGACAGG	6349
Oryza sativa Ser12Term CG-TAG	TCGCCCCTCGTCGCTCCTTCGCCACGGGTT AACCCGTGGCGAAGCAGCGACGACGCGCCGACCTGTCGGCGAT GCCGAAGCCGGTGGCCTAGGTGGCGAGCTGGGACGTGAGA GCCGACATGGTGGTGCACACGGTCGGCTGTTTAG	6350
	CGCCACCTAGGCCACCGGTGCGGTGTTTAG	6351
	CGGTGGCCTAGGTGGCG	6352
Naxy starch .	CGGCTCTCACCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTC GGCATCGCCGACAGGTAGGCGCGTCGTCGCTGCTTCGCCACGG GTTCCAGGGCCTCAAGCCCCGTAGCCCAGCCGG	6353
Oryza sativa Ser22Term FCG-TAG	CCGCTGGGCTACGGGGCTTGAGGCCCTGGAACCCGTGGCGAA GCAGCGACGACGCCCTACCTGTCGGCGATGCCGAAGCCGGT GCCGAGGTGGCGAGCTGGGACGTGAGAGCCG	6354
	CGACAGGT AGGCCGCCGT	6355
	ACGGCGCCTACCTGTCG	6356
Waxy starch GBSS	CCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCC GACAGGTCGGCGCCGTAGTCGCTGCTTCGCCACGGGTTCCAGGG CCTCAAGCCCCGTAGCCCAGCCGGCGGGGACGC	6357
Oryza sativa Ser25Term TCG-TAG	GCTCAAGCCCCGTAGCCCAGCCGCGCCCCCCCCGCCGCAGCCGCCGCCGCCGC	635
	GGCGCCGTAGTCGCTGC	635
	GCAGCGAC <u>T</u> ACGGCGCC	636
Waxy starch GBSS	CGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCCGAC AGGTCGGCGCGTCGTAGCTGCTTCGCCACGGGTTCCAGGGCCT CAAGCCCCGTAGCCCAGCCGGCGGGACGCATC	636
Oryza sativa Ser26Term TCG-TAG	GATGCGTCCCGCCGGCTGGGCTACGGGGCTTGAGGCCCTGGAA CCCGTGGCGAAGCAGC <u>T</u> ACGACGGCGCCGACCTGTCGGCGATGC CGAAGCCGGTGGCCGAGGTGGCGAGCTGGGACG	636
	GCCGTCGTAGCTGCCGAGGTGCCGAGGTGAGGTGAGGTAGGTGAGGAG	636
	GAAGCAGCTACGACGGC	636

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
Waxy starch GBSS Hordeum vulgare	GTCTCTCACTGCAGGTAGCCACACCCTGTGCGCGCGCGCCATGGC GGCTCTGGCCACGTCCTAGCTCGCCACCTCCGGCACCGTCCTCG GCGTCACCGACAGATTCCGGCGTCCAGGTTTTC	6365
GIn8Term CAG-TAG	GAAAACCTGGACGCCGGAATCTGTCGGTGACGCCGAGGACGGTG CCGGAGGTGGCGAGCTAGGCGCCAGAGCCGCCATGGCGC CGCGCACAGGGTGTGGCTACCTGCAGTGAGAGAC	6366
	CCACGTCC <u>T</u> AGCTCGCC	6367
	GGCGAGCT <u>A</u> GGACGTGG	6368
Waxy starch GBSS Hordeum vulgare	ATGGCGGCTCTGGCCACGTCCCAGCTCGCCACCTCCGGCACCGT CCTCGGCGTCACCGACTGATTCCGGCGTCCAGGTTTTCAGGGCCT CAGGCCCCGGAACCCGGCGATGCGGCGCTTG	6369
Arg21Term AGA-TGA	CAAGCGCCGCATCCGCCGGGTTCCGGGGCCTGAGGCCCTGAAAA CCTGGACGCCGGAATCAGTCGGTGACGCCGAGGACGGTGCCGG AGGTGGCGAGCTGGGACGTGCCAGAGCCGCCAT	6370
	TCACCGAC <u>T</u> GATTCCGG	6371
	CCGGAATC <u>A</u> GTCGGTGA	6372
Waxy starch GBSS Hordeum vulgare	CAGCTCGCCACCTCCGGCACCGTCCTCGGCGTCACCGACAGATT CCGGCGTCCAGGTTTTTAGGGCCTCAGGCCCCGGAACCCGGCGG ATGCGGCGCTTGGTATGAGGACTATCGGAGCAA	6373
GIn28Term CAG-TAG	TTGCTCCGATAGTCCTCATACCAAGCGCCGCATCCGCCGGGTTCC GGGGCCTGAGGCCCTAAAAACCTGGACGCCGGAATCTGTCGGTG ACGCCGAGGACGGTGCCGGAGGTGCCAGCTG	6374
	CAGGTTTT <u>T</u> AGGGCCTC	6375
	GAGGCCCT <u>A</u> AAAACCTG	6376
Waxy starch GBSS Hordeum vulgare	GGTTTTCAGGGCCTCAGGCCCCGGAACCCGGCGGATGCGGCGCT TGGTATGAGGACTATCTGAGCAAGCGCCGCCCCGAAGCAAAGCC GGAAAGCGCACCGCGGGAGCCGGCGGTGCCTCT	6377
Gly46Term GGA-TGA	AGAGGCACCGCCGGCTCCCGCGTGCGCTTTCCGGCTTTGCTTC GGGGCGGCGCTTGCTCAGATAGTCCTCATACCAAGCGCCGCATC CGCCGGGTTCCGGGGCCTGAGGCCCTGAAAACC	6378
	GGACTATC <u>T</u> GAGCAAGC	6379
	GCTTGCTC <u>A</u> GATAGTCC	6380
Waxy starch GBSS Hordeum vulgare	CCCCGGAACCCGGCGGATGCGGCGCTTGGTATGAGGACTATCGG AGCAAGCGCCGCCCCGTAGCAAAGCCGGAAAGCGCACCGCGGG AGCCGGCGGTGCCTCTCCGTGGTGAGCGCCA	6381
Lys52Term AAG-TAG	TGGCGCTCACCACCACGGAGAGGCACCGCCGGCTCCCGCGGTG CGCTTTCCGGCTTTGCTACGGGGCGCGCGCTTGCTCCGATAGTCC TCATACCAAGCGCCGCATCCGCCGGGTTCCGGGG	6382

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	CCGCCCGTAGCAAAGC	6383
	GCTTTGCTACGGGGCGG	6384
	ACGTCTTTCTCTCTCCTACGCAGTGGATTAATCGGCATGGCGG	6385
Vaxy starch	CTCTGGCCACGTCGTAGCTCGCAACGCGCCCGGCCTGGGC	
GBSS	GTCCCGGACGCGTCCACGTTCCGCCGCGCGCG	
Zea mays	CGCCGCGGAACGTGGACGCGTCCGGACGCCCAGGCCGGC	6386
SIn8Term	GCGCGTTGCGACGAGCTACGACGTGGCCAGAGCCGCCATGCCGA	
CAG-TAG	TTAATCCACTGCGTAGGAGAGAGAGAAAAGACGT	
	CCACGTCG <u>T</u> AGCTCGTC	6387
	GACGAGCT <u>A</u> CGACGTGG	6388
	GTCGCAACGCGCCCGGCCTGGGCGTCCCGGACGCGTCCACGT	6389
Naxy starch	TCCGCCGCGCGCGCGTAGGGCCTGAGGGGGGCCCGGGCGTC	
GBSS	LCCCCCCCCCCACACGCTCAGCATGCGGACCAGCG	
Zea mays	CCCTCCTCCCATGCTGAGCGTGTCCGCCGCCGCCGACGCCCG	6390
GIn30Term CAG-TAG	GGCCCCCTCAGGCCCTACGCGGCGCGCGCGGAACGTGGAC	
CAG-TAG	GCGTCCGGGACGCCCAGGCCGCGCGCGCGTTGCGAC	
	GCGCCGCGTAGGGCCTG	6391
	CAGGCCCTACGCGGCGC	6392
Morer storoh	TCCCGGACGCGTCCACGTTCCGCCGCGCGCGCGCGCGCAGGGCCT	6393
Waxy starch GBSS	GAGGGGGGCCCGGGCGTAGGCGGCGGCGGCGGCGCTCAGCATG	
Zea mays	LCCACCACCACCACCACCACCACCACCACCACCACCACCA	
Ser38Term	TECTEGTECCTEGECCCCCCCCCCCCCCCCCCCCCCCCC	639
TCG-TAG	IGCGTGTCCGCCGCCGCCTACGCCCGGGCCCCCTCAGGCCCTG	
100 17.0	CGCGCCGCGCGGAACGTGGACGCGTCCGGGA	
	CCGGGCGT <u>A</u> GGCGGCGG	639
	CCGCCGCCTACGCCCGG	639
Waxy starch	GCGTCGGCGGCGGGCGGACACGCTCAGCATGCGGACCAGCGCGC	639
GBSS	LCCCCGCCCCAGGCACTAGCAGCAGGCGCGCGCGCGGGGGCA	Ì
Zea mays	LCCTTCCCGTCGTCGTCGTGTGCGCCAGCGCUGGUA	000
Ser57Term	TGCCGCGCTGGCGCACACGACGAGCGACGGGAACCTGCCCCC	639
CAG-TAG	lecencene Technique (Control of the Control of the	
	GTCCGCATGCTGAGCGTGTCCGCCGCCGACGC	639
	CCAGGCAC <u>T</u> AGCAGCAG	640
1 .	CTGCTGCTAGTGCCTGG	040

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch	TCGGCGGCGGCGACACGCTCAGCATGCGGACCAGCGCGCGC	6401
GBSS	CGGCGCCCAGGCACCAG <u>T</u> AGCAGGCGCGCGCGGGGGGCAGGTT	
Zea mays	CCCGTCGCTCGTGTGCGCCAGCGCCGGCATGA	
GIn58Term	TCATGCCGGCGCTGGCGCACACGACGACGACGGGAACCTGCCC	6402
CAG-TAG	CCGCGGCGCGCTGCTACTGGTGCCTGGGCGCGCGCGCGCG	
	TGGTCCGCATGCTGAGCGTGTCCGCCGCCGCCGA	
	GGCACCAG <u>T</u> AGCAGGCG	6403
	CGCCTGCT <u>A</u> CTGGTGCC	6404

Example 11 <u>Altering fatty acid content of plants</u>

Improved means to manipulate fatty acid compositions, from biosynthetic or natural plant sources, are needed. For example, oils containing reduced saturated fatty acids are desired for dietary reasons and oils containing increased saturated fatty acids are also needed as alternatives to current sources of highly saturated oil products, such as tropical oils or chemically hydrogenated oils. It would therefore be advantageous to influence directly the production and composition of fatty acids in crop plants.

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Higher plants synthesize fatty acids, primarily palmitic, stearic and oleic acids, in the plastids (i.e., chloroplasts, proplastids, or other related organelles) as part of the Fatty Acid Synthase (FAS) complex. Fatty acid synthesis is the result of the three enzymatic activities: acyl-ACP elongase, acyl-ACP desaturase and acyl-ACP thioesterases specific for each of palmitoyl-, stearoyl- and oleoyl-ACP.

A variety of enzymes have been identified that influence the relative levels of saturated vs. unsaturated fatty acids in plants. For example, the enzymes stearoyl-acyl carrier protein (stearoyl-ACP) desaturase, oleoyl desaturase and linoleate desaturase produce unsaturated fatty acids from saturated precursors. Similarly, relative enzymatic activities of the various acyl-ACP thioesterases influences the relative acyl-chain composition of the resultant fatty acids. Consequently a reduction or an increase of the activity of these enzymes can alter the properties of oils produced in a plant. In fact, specific targeting of particular enzymatic activities can results in altered levels of particular fatty acids.

The attached tables disclose exemplary oligonucleotides base sequences which can be used to generate site-specific mutations in plant genes encoding proteins involved in fatty acid biosynthesis.

Table 22
Oligonucleotides to produce plants with reduced palmitate

Phenotype, Gene, Plant & Targeted	Altering Øligos	SEQ ID NO:
Acyl-ACP-thioesterase	TTTGGTGGCAGTGTCTTTGAACGCTTCATCTCCTCGTCATGGTGGC CACCTCTGCTACGTAGTCATTCTTTCCTGTACCATCTTCTTCACTT GATCCTAATGGAAAAGGCAATAAGATTGG	6405
Ser8Term	CCAATCTTATTGCCTTTTCCATTAGATTCC CCAATCTTATTGCCTTTTCCATTAGGATCAAGTGAAGAAGATGGTA CAGGAAAGAATGAC <u>T</u> ACGTAGCAGAGGTGGCCACCATGACGAGG AGATGAAGCGTTCAAAGACACTGCCACCAAA	6406

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGCTACGT <u>A</u> GTCATTCT	6407
	AGAATGAC <u>T</u> ACGTAGCA	6408
Reduced palmitate Acyl-ACP-thioesterase Arabidopsis thaliana	GGTGGCAGTGTCTTTGAACGCTTCATCTCCTCGTCATGGTGGCCA CCTCTGCTACGTCGTGATTCTTTCCTGTACCATCTTCTTCACTTGAT CCTAATGGAAAAGGCAATAAGATTGGGTC	6409
Ser9Term TCA-TGA	GACCCAATCTTATTGCCTTTTCCATTAGGATCAAGTGAAGAAGATG GTACAGGAAAGAAT <u>C</u> ACGACGTAGCAGAGGTGGCCACCATGACGA GGAGATGAAGCGTTCAAAGACACTGCCACC	6410
	TACGTCGT <u>G</u> ATTCTTTC	6411
	GAAAGAAT <u>C</u> ACGACGTA	6412
Reduced palmitate Acyl-ACP-thioesterase Arabidopsis thaliana	ATCTCCTCGTCATGGTGGCCACCTCTGCTACGTCGTCATTCTTTCC TGTACCATCTTCTTGACTTGA	6413
Ser17Term TCA-TGA	GAATTGAGTCCAGCAAGATTCGTAGACCCAATCTTATTGCCTTTTC CATTAGGATCAAGTCAAG	6414
	ATCTTCTT <u>G</u> ACTTGATC	6415
	GATCAAGT <u>C</u> AAGAAGAT	6416
Reduced palmitate Acyl-ACP-thioesterase <i>Arabidopsis thaliana</i>	GTGGCCACCTCTGCTACGTCGTCATTCTTTCCTGTACCATCTTCTT CACTTGATCCTAATTGAAAAGGCAATAAGATTGGGTCTACGAATCT TGCTGGACTCAATTCTGCACCTAACTCTG	6417
Gly22Term GGA-TGA	CAGAGTTAGGTGCAGAATTGAGTCCAGCAAGATTCGTAGACCCAA TCTTATTGCCTTTTCAATTAGGATCAAGTGAAGAAGATGGTACAGG AAAGAATGACGACGTAGCAGAGGTGGCCAC	6418
	ATCCTAAT <u>T</u> GAAAAGGC	6419
	GCCTTTTC <u>A</u> ATTAGGAT	6420
Reduced palmitate Acyl-ACP-thioesterase Garcinia mangostana	GCTTGAATTTGTGATCGATTGGTTAATTGTGGCCACAATGGTTGC TACTGCCGCCACGTGATCATTCTTTCCGTTGACTTCCCCTTCTGGG GATGCCAAATCGGGCAATCCCGGAAAAGG	6421
Ser8Term TCA-TGA	CCTTTTCCGGGATTGCCCGATTTGGCATCCCCAGAAGGGGAAGTC AACGGAAAGAATGATCACGCGCGCAGTAGCAACCATTGTGGCC ACAATTAACCAATCAGATCACAAATTCAAGC	6422
	CGCCACGT <u>G</u> ATCATTCT	6423
	AGAATGAT <u>C</u> ACGTGGCG	6424

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Reduced palmitate	TGAATTTGTGATCTGATTGGTTAATTGTGGCCACAATGGTTGCTAC TGCCGCCACGTCATGATTCTTTCCGTTGACTTCCCCTTCTGGGGAT GCCAAATCGGGCAATCCCGGAAAAGGGTC	6425
Garcinia mangostana Ger9Term CA-TGA	GCCAAATCGGGCAATCCCGGAAACGGTC GACCCTTTTCCGGGATTGCCCGATTTGGCATCCCCAGAAGGGGAA GTCAACGGAAAGAATCATGACGTGGCGGCAGTAGCAACCATTGTG GCCACAATTAACCAATCAGATCACAAATTCA	6426
	CACGTCATGACCATTCACCATTCACCATTCACCACTCACT	6427
	GAAAGAAT <u>C</u> ATGACGTG	6428
Reduced palmitate Acyl-ACP-thioesterase Garcinia mangostana	CTGATTGGTTAATTGTGGCCACAATGGTTGCTACTGCCGCCACGT CATCATTCTTTCCGTAGACTTCCCCTTCTGGGGATGCCAAATCGG GCAATCCCGGAAAAGGGTCGGTGAGTTTTGG	6429
Leu13Term TTG-TAG	CCAAAACTCACCGACCCTTTTCCGGGATTGCCCGATTTGGCATCC CCAGAAGGGGAAGTCTACGGAAAGAATGATGACGTGGCGGCAGT AGCAACCATTGTGGCCACAATTAACCAATCAG	6430
	CTTTCCGTAGACTTCCC	6431
	GGGAAGTC <u>T</u> ACGGAAAG	6432
Reduced palmitate Acyl-ACP-thioesterase	ATGGTTGCTACTGCCGCCACGTCATCATTCTTTCCGTTGACTTCCC CTTCTGGGGATGCCTAATCGGGCAATCCCGGAAAAGGGTCGGTG AGTTTTGGGTCAATGAAGTCGAAATCCGCGG	6433
Garcinia mangostana Lys21Term AAA-TAA	CCGCGGATTTCGACTTCATTGACCCAAAACTCACCGACCCTTTTCC GGGATTGCCCGATTAGGCATCCCCAGAAGGGGAAGTCAACGGAA AGAATGATGACGTGGCGGCAGTAGCAACCAT	6434
	GGGATGCCTAATCGGCC	6435
	GCCCGATTAGGCATCCC	6436
Reduced palmitate Acyl-ACP-thioesterase	GGGATTICAGCACGAAATTGAAGTTGTTTTAAAAACCATGGTTGC	6437
Gossypium hirsutum Ser8Term TCG-TAG	CCGAGCTTCTTGTTTTTCGAGTCAGAGGAGTCAGGTGAAGAAGTG ACTGGGAAAAACGCCTATGTCACAGCAGTAGCAACCATGGTTTTTA	643
	AAAACAACTTCAATTTCGTGCTGAAATCCC TGTGACAT <u>A</u> GGCGTTTT	643
	AAAACGCC <u>T</u> ATGTCACA	644
Reduced palmitate Acyl-ACP-thioesterase	TGTTTTTAAAAACCATGGTTGCTACTGCTGTGACATCGGCGTTTTT CCCAGTCACTTCTTGACCTGACTCGAAAAACAAGAAG CTCGGAAGCATCAAGTCGAAGCCATCGGT	L
Gossypium hirsutum Ser16Term TCA-TGA	ACCGATGCTTCGACTTCGACTTCCGAGCTTCTTGTTTTTCGAGT CAGAGGAGTCAGGTCAAGAAGTGACTGGGAAAAACGCCGATGTCA CAGCAGTAGCAACCATGGTTTTTAAAAACA	644

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CACTTCTT G ACCTGACT	6443
	AGTCAGGT <u>C</u> AAGAAGTG	6444
Reduced palmitate Acyl-ACP-thioesterase Gossypium hirsutum	TTGCTACTGCTGTGACATCGGCGTTTTTCCCAGTCACTTCTTCACC TGACTCCTCTGACTAGAAAAAAAAAA	6445
Ser22Term TCG-TAG	TGCAAACTTCCAGAAGAAACCGATGGCTTCGACTTGATGCTTCCGA GCTTCTTGTTTTTCTAGTCAGAGGAGTCAGGTGAAGAAGTGACTGG GAAAAACGCCGATGTCACAGCAGTAGCAA	6446
	CTCTGACT <u>A</u> GAAAACA	6447
	TGTTTTC <u>T</u> AGTCAGAG	6448
Reduced palmitate Acyl-ACP-thioesterase Gossypium hirsutum	GCTACTGCTGTGACATCGGCGTTTTTCCCAGTCACTTCTTCACCTG ACTCCTCTGACTCGTAAAACAAGAAGCTCGGAAGCATCAAGTCGA AGCCATCGGTTTCTTCTGGAAGTTTGCAAG	6449
Lys23Term AAA-TAA	CTTGCAAACTTCCAGAAGAAACCGATGGCTTCGACTTGATGCTTCC GAGCTTCTTGTTTTACGAGTCAGAGGAGTCAGGTGAAGAAGTGAC TGGGAAAAACGCCGATGTCACAGCAGTAGC	6450
	CTGACTCG <u>T</u> AAAACAAG	6451
	CTTGTTTT <u>A</u> CGAGTCAG	6452
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	CTCCCGCTCGTTGAAAGACAATGGTGGCTACCGCTGCAAGCTCTG CATTCTTCCCCGTGTAGTCCCCGGTCACCTCCTCTAGACCAGGAA AGCCCGGAAATGGGTCATCGAGCTTCAGCCC	6453
Ser14Term TCG-TAG	GGGCTGAAGCTCGATGACCCATTTCCGGGCTTTCCTGGTCTAGAG GAGGTGACCGGGGAC <u>T</u> ACACGGGGAAGAATGCAGAGCTTGCAGC GGTAGCCACCATTGTCTTTCAACGAGCGGGAG	6454
	CCCCGTGT <u>A</u> GTCCCCGG	6455
	CCGGGGAC <u>T</u> ACACGGGG	6456
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	ATGGTGGCTACCGCTGCAAGCTCTGCATTCTTCCCCGTGTCGTCC CCGGTCACCTCCTTGACCAGGAAAGCCCGGAAATGGGTCATCG AGCTTCAGCCCCATCAAGCCCAAATTTGTCG	6457
Arg21Term AGA-TGA	CGACAAATTTGGGCTTGATGGGGCTGAAGCTCGATGACCCATTTC CGGGCTTTCCTGGTC <u>A</u> AGAGGAGGTGACCGGGGACGACACGGG GAAGAATGCAGAGCTTGCAGCGGTAGCCACCAT	6458
	CCTCCTCT <u>T</u> GACCAGGA	6459
	TCCTGGTC <u>A</u> AGAGGAGG	6460

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Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
avi ACD thinesterase	ACCTCCTCTAGACCATGAAAGCCCGGAAATGGGTCATCGAGCTTC	6461
Cuphea hookeriana Gly23Term GGA-TGA	AGCCCCATCAAGCCCAAATTTGTCGCCAATG CATTGGCGACAAATTTGGGCTTGATGGGGCTGAAGCTCGATGACC CATTTCCGGGCTTTCATGGTCTAGAGGAGGTGACCGGGGACGAC ACGGGGAAGAATGCAGAGCTTGCAGCGGTAGC	6462
	CTAGACCATGAAAGCCC	6463
	GGGCTTTCATGGTCTAG	6464
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	ACCGCTGCAAGCTCTGCATTCTTCCCCGTGTCGTCCCCGGTCACC TCCTCTAGACCAGGATAGCCCGGAAATGGGTCATCGAGCTTCAGC CCCATCAAGCCCAAATTTGTCGCCAATGGCG	6465
_ys24Term AAG-TAG	CGCCATTGGCGACAAATTTGGGGCTTGATGGGGCTGAAGCTCGATG ACCCATTTCCGGGCTATCCTGGTCTAGAGGAGGTGACCGGGGAC GACACGGGGAAGAATGCAGAGCTTGCAGCGGT	6466
	GACCAGGA <u>T</u> AGCCCGGA	6467
٦	TCCGGGCT <u>A</u> TCCTGGTC	6468
Reduced palmitate Acyl-ACP-thioesterase	IGCCACCGCTGCAAGTTCTGCATTCTTCCCCCTGCCGTCCCCGGAC ACCTCCTCTAGGCCGTGAAAGCTCGGAAATTTGTCGCCAATG	6469
Cuphea lanceolata Gly23Term GGA-TGA	CATTGGCGACAAATTTGGGCTTGAGGGGGGCTCAAGCTCGATGACC	6470
	AGGGGAAGAATGCAGAACTTGCAGCGGTGGC CTAGGCCGTGAAAGCTC	6471
	GAGCTTTCACGGCCTAG	6472
Reduced palmitate Acyl-ACP-thioesterase	ACCGCTGCAAGTTCTGCATTCTTCCCCCTGCCGTCCCCGGACACC	6473
Cuphea lanceolata Lys24Term AAG-TAG	CGCCTCAAGCCCAAATTTGTCGGGTTTGCGGGGGGCTCAAGCTCGAT CGGCATTGCGACCTATCCCGGCCTAGAGGAGGTGTCCGGGGA CGGCAGGGGGAAGAATGCAGAACTTGCAGCGGT	647
	GGCCGGGATAGCTCGGA	647
	TCCGAGCTATCCCGGCC	647
Reduced palmitate Acyl-ACP-thioesterase Cuphea lanceolata Gly26Term GGA-TGA	IOAACCCCAAATTCTCCCCAAIGCCGGGIIGA	
	TCAACCCGGCATTGTGGGGACAAATTTGGGCTTGAGGGGGGCTCAAGC TCGATGACCCATTTCAGAGCTTTCCCGGCCTAGAGGAGGTGTCCG GGGACGGCAGGGGAAGAATGCAGAACTTGC	647

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GAAAGCTC <u>T</u> GAAATGGG	6479
	CCCATTTC <u>A</u> GAGCTTTC	6480
Reduced palmitate Acyl-ACP-thioesterase Cuphea lanceolata	CATTCTTCCCCCTGCCGTCCCCGGACACCTCCTCTAGGCCGGGAA AGCTCGGAAATGGGTGATCGAGCTTGAGCCCCCTCAAGCCCAAAT TTGTCGCCAATGCCGGGTTGAAGGTTAAGGC	6481
Ser29Term TCA-TGA	GCCTTAACCTTCAACCCGGCATTGGCGACAAATTTGGGCTTGAGG GGGCTCAAGCTCGATCACCCATTTCCGAGCTTTCCCGGCCTAGAG GAGGTGTCCGGGGACGGCAGGGGGAAGAATG	6482
	AAATGGGT <u>G</u> ATCGAGCT	6483
	AGCTCGAT <u>C</u> ACCCATTT	6484
Reduced palmitate Acyl-ACP-thioesterase Helianthus annuus	CGTTTAAGTGGATCGGACATTTAAGTGTTTTAATCATGGTAGCTAT GAGTGCTACTGCGT A GCTGTTTCCGGTTTCTTCCCCAAAACCTCA CTCTGGAGCCAAGACATCTGATAAGCTTGG	6485
Ser9Term TCG-TAG	CCAAGCTTATCAGATGTCTTGGCTCCAGAGTGAGGTTTTGGGGAA GAAACCGGAAACAGC <u>T</u> ACGCAGTAGCACTCATAGCTACCATGATT AAAACACTTAAATGTCCGATCCACTTAAACG	6486
	TACTGCGTAGCTGTTTC	6487
	GAAACAGC <u>T</u> ACGCAGTA	6488
Reduced palmitate Acyl-ACP-thioesterase Helianthus annuus	AGTGTTTTAATCATGGTAGCTATGAGTGCTACTGCGTCGCTGTTTC CGGTTTCTTCCCCATAACCTCACTCTGGAGCCAAGACATCTGATAA GCTTGGAGGTGAACCAGGTAGTGTTGCTG	6489
Lys17Term AAA-TAA	CAGCAACACTACCTGGTTCACCTCCAAGCTTATCAGATGTCTTGGC TCCAGAGTGAGGTTATGGGGGAAGAACCGGAAACAGCGACGCAG TAGCACTCATAGCTACCATGATTAAAACACT	6490
	CTTCCCCA <u>T</u> AACCTCAC	6491
	GTGAGGTT <u>A</u> TGGGGAAG	6492
Reduced palmitate Acyl-ACP-thioesterase Helianthus annuus	ATGGTAGCTATGAGTGCTACTGCGTCGCTGTTTCCGGTTTCTTCCC CAAAACCTCACTCTTGAGCCAAGACATCTGATAAGCTTGGAGGTGA ACCAGGTAGTGTTGCTGTGCGCGGAATCA	6493
Gly21Term GGA-TGA	TGATTCCGCGCACAGCAACACTACCTGGTTCACCTCCAAGCTTATC AGATGTCTTGGCTCAAGAGTGAGGTTTTGGGGAAGAAACCGGAAA CAGCGACGCAGTAGCACTCATAGCTACCAT	6494
	CTCACTCT <u>T</u> GAGCCAAG	6495
	CTTGGCTC A AGAGTGAG	6496

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
cyl-ACP-thioesterase	GCTATGAGTGCTACTGCGTCGCTGTTTCCGGTTTCTTCCCCAAAAC CTCACTCTGGAGCCTAGACATCTGATAAGCTTGGAGGTGAACCAG GTAGTGTTGCTGTGCGCGGAATCAAGACAA	6497
lelianthus annuus ys23Term AG-TAG	TTGTCTTGATTCCGCGCACACCACCTACCTGGTTCACCTCCAAG CTTATCAGATGTCTAGGCTCCAGAGTGAGGTTTTGGGGAAGAAAC CGGAAACAGCGACGCAGTAGCACTCATAGC	6498
	CTGGAGCC <u>T</u> AGACATCT	6499
	AGATGTCT <u>A</u> GGCTCCAG	6500
Reduced palmitate Acyl-ACP-thioesterase Cuphea palustris	ATGGTGGCTGCAGCAAGTTCTGCATGCTTCCCTGTTCCATCC CCAGGAGCCTCCCCTTAACCTGGGAAGTTAGGCAACTGGTCATCG AGTTTGAGCCCTTCCTTGAAGCCCAAGTCAA	6501
_ys21Term AAA-TAA	TTGACTTGGGCTTCAAGGAAGGGCTCAAACTCGATGACCAGTTGC CTAACTTCCCAGGTTAAGGGGAGGCTCCTGGGGATGGAACAGGG AAGCATGCAGAACTTGCTGCAGCAGCCACCAT	6502
	CCTCCCCTTAACCTGGG	6503
	CCCAGGTT A AGGGGAGG	6504
Reduced palmitate Acyl-ACP-thioesterase	ICCTTCCTTGAAGCCCAAGIUAATUUUCAATUU	6505
Cuphea palustris Lys24Term AAG-TAG	CATTGGGGATTGACTTGGGCTTCAAGGAAGGGCTCAAACTCGATG ACCAGTTGCCTAACTACCCAGGTTTAGGGGAGGCTCCTGGGGATG GAACAGGGAAGCATGCAGAACTTGCTGCAGC	6506
	AACCTGGGTAGTTAGGC	650
	GCCTAACTACCCAGGTT	650
Reduced palmitate Acyl-ACP-thioesterase	TGCATGCTTCCCTGTTCCATCCCCAGGAGCCTCCCTGAACCTGG	650
Cuphea palustris Trp28Term TGG-TGA	AACCTGAAATCCGCCATTGGGGATTGACTTGGGCTTCAAGGAAGG	651
	GGCTCCTGGGGATGGAACAGGGAAGCATGCA GGCAACTGATCATCGAG	651
	CTCGATGA <u>T</u> CAGTTGCC	651
Reduced palmitate Acyl-ACP-thioesteras Cuphea palustris	LACTCAATCCCCAATGGCGGALLICAGGLIAA	
Ser29Term TCA-TGA	TTAACCTGAAATCCGCCATTGGGGATTGACTTGGGCTTCAAGGAA GGGCTCAAACTCGATCACCAGTTGCCTAACTTCCCAGGTTTAGGG GAGGCTCCTGGGGATGGAACAGGGAAGCATG	65

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CAACTGGT <u>G</u> ATCGAGTT	6515
	AACTCGAT <u>C</u> ACCAGTTG	6516
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	ATGGTGGCTGCCGCAGCAAGTTCTGCATTCTTCTCCGTTCCAACC CCGGGAATCTCCCCTTAACCCGGGAAGTTCGGTAATGGTGGCTTT CAGGTTAAGGCAAACGCCAATGCCCATCCTA	6517
Lys21Term AAA-TAA	TAGGATGGCATTGGCGTTTGCCTTAACCTGAAAGCCACCATTAC CGAACTTCCCGGGTT <u>A</u> AGGGGAGATTCCCGGGGTTGGAACGGAG AAGAATGCAGAACTTGCTGCGGCAGCCACCAT	6518
·	TCTCCCCT <u>T</u> AACCCGGG	6519
	CCCGGGTT <u>A</u> AGGGGAGA	6520
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	GCCGCAGCAAGTTCTGCATTCTTCTCCGTTCCAACCCCGGGAATC TCCCCTAAACCCGGG <u>T</u> AGTTCGGTAATGGTGGCTTTCAGGTTAAG GCAAACGCCAATGCCCATCCTAGTCTAAAGT	6521
Lys24Term AAG-TAG	ACTITAGACTAGGATGGGCATTGGCGTTTGCCTTAACCTGAAAGCC ACCATTACCGAACT <u>A</u> CCCGGGTTTAGGGGAGATTCCCGGGGTTGG AACGGAGAAGAATGCAGAACTTGCTGCGGC	6522
	AACCCGGG <u>T</u> AGTTCGGT	6523
	ACCGAACT <u>A</u> CCCGGGTT	6524
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	TTCTCCGTTCCAACCCCGGGAATCTCCCCTAAACCCGGGAAGTTC GGTAATGGTGGCTTT <u>T</u> AGGTTAAGGCAAACGCCAATGCCCATCCT AGTCTAAAGTCTGGCAGCCTCGAGACTGAAG	6525
Gln31Term CAG-TAG	CTTCAGTCTCGAGGCTGCCAGACTTTAGACTAGGATGGCATTGG CGTTTGCCTTAACCT <u>A</u> AAAGCCACCATTACCGAACTTCCCGGGTTT AGGGGAGATTCCCGGGGTTGGAACGGAGAA	6526
	GTGGCTTT <u>T</u> AGGTTAAG	6527
	CTTAACCT <u>A</u> AAAGCCAC	6528
Reduced palmitate Acyl-ACP-thioesterase Cuphea hookeriana	GTTCCAACCCGGGAATCTCCCCTAAACCCGGGAAGTTCGGTAAT GGTGGCTTTCAGGTT <u>T</u> AGGCAAACGCCAATGCCCATCCTAGTCTA AAGTCTGGCAGCCTCGAGACTGAAGATGACA	6529
Lys33Term AAG-TAG	TGTCATCTTCAGTCTCGAGGCTGCCAGACTTTAGACTAGGATGGG CATTGGCGTTTGCCT <u>A</u> AACCTGAAAGCCACCATTACCGAACTTCCC GGGTTTAGGGGAGATTCCCGGGGTTGGAAC	6530
	TTCAGGTT <u>T</u> AGGCAAAC	6531
	GTTTGCCT <u>A</u> AACCTGAA	6532

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
educed palmitate	ATGTTGAAGCTCTCGTGTAATGCGACTGATAAGTTACAGACCCTCT	6533
cyl-ACP-thioesterase	TCTCGCATTCTCATTAACCGGATCCGGCACACCGGAGAACCGTCT CCTCCGTGTCGTGCTCTCATCTGAGGAAAC	
rassica rapa	GTTTCCTCAGATGAGAGCACGACACGGAGAGACGGTTCTCCGGT	6534
Gin21Term CAA-TAA	GTGCCGGATCCGGTTAATGAGAATGCGAGAAGAGGGTCTGTAACT	
,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	TATCAGTCGCATTACACGAGAGCTTCAACAT	CESE
	ATTCTCAT <u>T</u> AACCGGAT	6535
	ATCCGGTT <u>A</u> ATGAGAAT	6536
Reduced palmitate	GCGACTGATAAGTTACAGACCCTCTTCTCGCATTCTCATCATCATCATCATCATCATCATCATCATCATC	6537
Acyl-ACP-thioesterase	ATCCGGCACACCGGTGAACCGTCTCCTCCGTGTCGTGCTCTCATC	
Brassica rapa	TGAGGAAACCGGTTCTCGATCCTTTGCGAG	6538
Arg28Term	CTCGCAAAGGATCGAGAACCGGTTTCCTCAGATGAGAGCACGACA	0000
AGA-TGA	CGGAGGAGACGGTTCACCGGTGTGCCGGATCCGGTTGATGAGAA TGCGAGAAGAGGGTCTGTAACTTATCAGTCGC	
	CACACCGGTGAACCGTC	6539
	GACGGTTCACCGGTGTG	6540
	CCCTCTTCTCGCATTCTCATCAACCGGATCCGGCACACCGGAGAA	6541
Reduced palmitate		
Acyl-ACP-thioesterase Brassica rapa	LATCCTTTGCGAGCGATCGTATCTGCTGATCA	
Ser24Term	TGATCAGCAGATACGATCGCTCGCAAAGGATCGAGAACCGGIIIC	6542
TCG-TAG	CTCAGATGAGAGCACTACACGGAGGAGACGGTTCTCCGGTGTGC	
	CGGATCCGGTTGATGAGAATGCGAGAAGAGGG	6543
	CTCCGTGTAGTGCTCTC	
	GAGAGCAC <u>T</u> ACACGGAG	654
Reduced palmitate	CTTCTCGCATTCTCATCAACCGGATCCGGCACACCGGAGAACCGT	654
Acyl-ACP-thioesterase	CTCCTCCGTGTCGTGATCTCATCTGAGGAAACCGGTTCTCGATCC	
Brassica rapa	TTTGCGAGCGATCGTATCTGCTGATCAAGGA	654
Cys25Term	TCCTTGATCAGCAGATACGATCGCTCGCAAAGGATCGAGAACCGG	057
TGC-TGA	TTTCCTCAGATGAGATCACGACACGGAGGAGGAGGGTTCTCCGGTG	
	TGCCGGATCCGGTTGATGAGAATGCGAGAAG GTGTCGTGATCTCATCT	654
		654
	AGATGAGA <u>T</u> CACGACAC IATTCTTCTATAAACCAAAACCTCAGGAACCATAAAAAAAA	654
Reduced palmitate Acyl-ACP-thioesterase Brassica napus Leu2Term TTG-TAG		
	ACACCTTCTCCTTCTTCTCCGATTCCTC	
	GAGGAATCGGAGAAGAAGGAGAAGGTGTGTAAGTTGTTAGTCACA	655
	TTACACGAAAGCTTCTACATTTTTGATGCCCTTTTTTTTT	
	CTGAGGTTTTGGTTTATAGAAGAAGAAT	<u></u>

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	AAAAATGT <u>A</u> GAAGCTTT	6551
	AAAGCTTC <u>T</u> ACATTTTT	6552
Reduced palmitate Acyl-ACP-thioesterase Brassica napus	TCTTCTTCTATAAACCAAAACCTCAGGAACCATAAAAAAAA	6553
Lys3Term AAG-TAG	GGGAGGAATCGGAGAAGAAGGAGAAGGTGTGAAGTTGTTAGTCA CATTACACGAAAGCT <u>A</u> CAACATTTTTGATGCCCTTTTTTTTTATGG TTCCTGAGGTTTTGGTTTATAGAAGAAGA	6554
	AAATGTTG <u>T</u> AGCTTTCG	6555
	CGAAAGCT <u>A</u> CAACATTT	6556
Reduced palmitate Acyl-ACP-thioesterase Brassica napus Ser5Term TCG-TAG	CTATAAACCAAAACCTCAGGAACCATAAAAAAAAAAAGGGCATCAAA AATGTTGAAGCTTT <u>A</u> GTGTAATGTGACTAACAACTTACACACCTTCT CCTTCTTCTCCGATTCCTCCCTTTTCAT	6557
	ATGAAAAGGGAGGAATCGGAGAAGAAGGAGAAGGTGTGAAGTTG TTAGTCACATTACAC <u>T</u> AAAGCTTCAACATTTTTGATGCCCTTTTTTTT TTATGGTTCCTGAGGTTTTGGTTTATAG	6558
	GAAGCTTT <u>A</u> GTGTAATG	6559
	CATTACAC <u>T</u> AAAGCTTC	6560
Reduced palmitate Acyl-ACP-thioesterase Brassica napus	AAACCAAAACCTCAGGAACCATAAAAAAAAAAAAGGGCATCAAAAATG TTGAAGCTTTCGTG <u>A</u> AATGTGACTAACAACTTACACACCTTCTCCTT CTTCTCCGATTCCTCCCTTTTCATCCCG	6561
Cys6Term TGT-TGA	CGGGATGAAAAGGGAGGAATCGGAGAAGAAGGAGAAGGTGTGTA AGTTGTTAGTCACATT <u>T</u> CACGAAAGCTTCAACATTTTTGATGCCCTT TTTTTTTTATGGTTCCTGAGGTTTTGGTTT	6562
	CTTTCGTG <u>A</u> AATGTGAC	6563
	GTCACATT <u>T</u> CACGAAAG	6564

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Table 23
Oligonucleotides to produce plants with increased stearate

.	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	Increased stearate stearoyl-ACP	GGGAGAGCTCTAGCTCTGTAGAAAAGAAGGATTCATTCAT	6565
0	desaturase Arabidopsis thaliana Lys4Term	TTGGCGGACGAGTCGAGGAAGGGAATTTGTAAGGCTGAGATGCCA CCAAAGGGTTAAACTATAGAGCCATTTCTGGATATGAATGA	6566
	AAG-TAG	TGGCTCTATAGTTTAAC	6567
		GTTAAACT <u>A</u> TAGAGCCA	6568
	Increased stearate stearoyl-ACP desaturase	CTCTGTAGAAAAGAAGGATTCATTCATCATATCCAGAAATGGCTCT AAAGTTTAACCCTTAGGTGGCATCTCAGCCTTACAAATTCCCTTCC TCGACTCGTCCGCCAACTCCTTCTTTCAG	6569
15	Arabidopsis thaliana Leu8Term TTG-TAG	CTGAAAGAAGGAGTTGGCGGACGAGTCGAGGAAGGGAATTTGTAA GGCTGAGATGCCACCTAAGGGTTAAACTTTAGAGCCATTTCTGGAT ATGATGAATGAATCCTTCTTTTCTACAGAG	6570
	TIG-TAG	TAACCCTT <u>A</u> GGTGGCAT	6571
		ATGCCACC <u>T</u> AAGGGTTA	6572
00	Increased stearate stearoyl-ACP	AGAAGGATTCATTCATCATATCCAGAAATGGCTCTAAAGTTTAACC CTTTGGTGGCATCTTAGCCTTACAAATTCCCTTCCTCGACTCGTCC GCCAACTCCTTCTTTCAGATCTCCCAAGT	6573
20	desaturase Arabidopsis thaliana Gln12Term	ACTTGGGAGATCTGAAAGAAGGAGTTGGCGGACGAGTCGAGGAA GGGAATTTGTAAGGCTAAGATGCCACCAAAGGGTTAAACTTTAGAG CCATTTCTGGATATGAATGAATCCTTCT	6574
	CAG-TAG	TGGCATCTTAGCCTTAC	6575
		GTAAGGCT <u>A</u> AGATGCCA	6576
25	Increased stearate stearoyl-ACP	TCATTCATCATATCCAGAAATGGCTCTAAAGTTTAACCCTTTGGTG GCATCTCAGCCTTAGAAATTCCCTTCCTCGACTCGTCCGCCAACTC CTTCTTTCAGATCTCCCAAGTTCCTCTGC	
	desaturase Arabidopsis thaliana Phe14Term	GCAGAGGAACTTGGGAGATCTGAAAGAAGGAGTTGGCGGACGAG TCGAGGAAGGGAATTTCTAAGGCTGAGATGCCACCAAAGGGTTAA ACTTTAGAGCCATTTCTGGATATGATGAATGA	6578
	TAC-TAG	CAGCCTTAGAGGGATTTGTGGATTGTGGATTGTGGATTGTGGATTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGTGGATTGTGGATTGTGTGGATTGTGTGGATTGTGTGTGATTGTGTGGATTGTGTGGATTGTGTGATTGTGTGGATTGTGTGATTGTGTGATTGTGTGATTGTGTGATTGTGTGATTGTGTGATTGTGTGATTGTGTGTGTGTGATTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGT	6579
		GGGAATTT <u>C</u> TAAGGCTG	6580

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	Increased stearate stearoyl-ACP desaturase	GAGAGCTCGCTCGTGTCTGAAAGAACATCAAACCTCGTATCAAAAA AAAGAAAATGGCAT <u>A</u> GAAGCTTAACCCTTTGGCATCTCAGCCTTAC AAACTCCCTTCCTCGGCTCGTCCGCCAAT	6581
5	Brassica napus Leu3Term TTG-TAG	ATTGGCGGACGAGCCGAGGAAGGGAGTTTGTAAGGCTGAGATGC CAAAGGGTTAAGCTTC <u>T</u> ATGCCATTTTCTTTTTTTGATACGAGGTT TGATGTTCTTTCAGACACGAGCGAGCTCTC	6582
		AATGGCAT <u>A</u> GAAGCTTA	6583
		TAAGCTTC <u>T</u> ATGCCATT	6584
	Increased stearate stearoyl-ACP desaturase	GAGCTCGCTCGTGTCTGAAAGAACATCAAACCTCGTATCAAAAAAA AGAAAATGGCATTGTAGCTTAACCCTTTGGCATCTCAGCCTTACAA ACTCCCTTCCTCGGCTCGTCCGCCAATCT	6585
10	Brassica napus Lys4Term AAG-TAG	AGATTGGCGGACGAGCCGAGGAAGGGAGTTTGTAAGGCTGAGAT GCCAAAGGGTTAAGCT <u>A</u> CAATGCCATTTTCTTTTTTTTGATACGAG GTTTGATGTTCTTTCAGACACGAGCGAGCTC	6586
		TGGCATTG <u>T</u> AGCTTAAC	6587
		GTTAAGCT <u>A</u> CAATGCCA	6588
15	Increased stearate stearoyl-ACP desaturase	TCTGAAAGAACATCAAACCTCGTATCAAAAAAAAAGAAAATGGCATT GAAGCTTAACCCTT <u>A</u> GGCATCTCAGCCTTACAAACTCCCTTCCTCG GCTCGTCCGCCAATCTCTACTCTCAGATC	6589
	Brassica napus Leu8Term TTG-TAG	GATCTGAGAGTAGAGATTGGCGGACGAGCCGAGGAAGGGAGTTT GTAAGGCTGAGATGCC <u>T</u> AAGGGTTAAGCTTCAATGCCATTTTCTTT TTTTTGATACGAGGTTTGATGTTCTTTCAGA	6590
		TAACCCTT <u>A</u> GGCATCTC	6591
		GAGATGCC <u>T</u> AAGGGTTA	6592
20	Increased stearate stearoyl-ACP desaturase	AACATCAAACCTCGTATCAAAAAAAAAGAAAATGGCATTGAAGCTTAA CCCTTTGGCATCTTAGCCTTACAAACTCCCTTCCTCGGCTCGTCCG CCAATCTCTACTCTCAGATCTCCCAAGT	6593
	Brassica napus Gln11Term CAG-TAG	ACTTGGGAGATCTGAGAGTAGAGATTGGCGGACGAGCCGAGGAA GGGAGTTTGTAAGGCT <u>A</u> AGATGCCAAAGGGTTAAGCTTCAATGCC ATTTTCTTTTTTTGATACGAGGTTTGATGTT	6594
		TGGCATCT <u>T</u> AGCCTTAC	6595
	·	GTAAGGCT <u>A</u> AGATGCCA	6596
25	Increased stearate stearoyl-ACP desaturase	AACCAAAAGAAAAAGGTAAGAAAAAAAAAACAATGGCTCTCAAGCTCA ATCCTTTCCTT	6597
30	Ricinus communis Gln27Term CAA-TAA	ACTTAGGAGATCTGGTACTGGCCATTGGTGGAAGAGCGAAAGAAG GTAACTTTTGGGTTT <u>A</u> AGAAAGGAAAGGATTGAGCTTGAGAGCCAT TGTTTTTTTCTTACCTTTTTCTTTTGGTT	6598

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	TCCTTTCTTAAACCCAA	6599
	TTGGGTTT A AGAAAGGA	6600
ncreased stearate tearoyl-ACP	AAGAAAAAGTAAGAAAAAAAAAACAATGGCTCTCAAGCTCAATCCTT TCCTTTCTCAAACCTAAAAGTTACCTTCTTTCGCTCTTCCACCAATG GCCAGTACCAGATCTCCTAAGTTCTACA	6601
esaturase Ricinus communis GIn29Term	TGTAGAACTTAGGAGATCTGGTACTTGGTGGAAGAGCGA AAGAAGGTAACTTTTAGGTTGAGAAAGGAAAG	6602
CAA-TAA	CTCAAACC <u>T</u> AAAAGTTA	6603
	TAACTTTT A GGTTTGAG	6604
ncreased stearate stearoyl-ACP	AAAAAGGTAAGAAAAAAAAAACAATGGCTCTCAAGCTCAATCCTTTCC TTTCTCAAACCCAATAGTTACCTTCTTTCGCTCTTCCACCAATGGC CAGTACCAGATCTCCTAAGTTCTACATGG	6605
desaturase Ricinus communis Lys30Term	CCATGTAGAACTTAGGAGATCTGGTACTGGCCATTGGTGGAAGAG CGAAAGAAGGTAACTATTGGGTTTGAGAAAGGAAAG	6606
AAG-TAG	AAACCCAATAGTTACCT	6607
	AGGTAACT <u>A</u> TTGGGTTT	6608
Increased stearate stearoyl-ACP	TCTCAAACCCAAAAGTTACCTTCTTTCGCTCTTCCACCAATGGCCA GTACCAGATCTCCTTAGTTCTACATGGCCTCTACCCTCAAGTCTGG TTCTAAGGAAGTTGAGAATCTCAAGAAGC	6609
desaturase <i>Ricinus communis</i> Lys46Term AAG-TAG	GCTTCTTGAGATTCTCAACTTCCTTAGAACCAGACTTGAGGGTAGA GGCCATGTAGAACTAAGGAGATCTGGTACTGGCCATTGGTGGAAG AGCGAAAGAAGGTAACTTTTGGGTTTGAGA	661
AAG-1AG	GATCTCCT <u>T</u> AGTTCTAC	661
	GTAGAACT A AGGAGATC	661
Increased stearate stearoyl-ACP desaturase	TCTTCTGATTCATTTAATCTTTACTCATCAATGGCTCTGAGACTGAA CCCTATCCCCACCTAAACCTTCTCCCTCCCCCAAATGGCCAGTCT CAGATCTCCCAGGTTCCGCATGGCCTCTA	661
Glycine max Gln11Term	TAGAGGCCATGCGGAACCTGGGAGATCTGAGACTGGCCATTTGG GGGAGGGAGAAGGTTTAGGTGGGGATAGGGTTCAGTCTCAGAGC CATTGATGAGTAAAGATTAAATGAATCAGAAGA	661
CAA-TAA	TCCCCACCTAAACCTTC	661
	GAAGGTTTAGGTGGGGA	661

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	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	Increased stearate stearoyl-ACP desaturase	CTTTACTCATCAATGGCTCTGAGACTGAACCCTATCCCCACCCA	6617
5	Glycine max Gln17Term CAA-TAA	TGGAACCGGAGCGAGGGTAGAGGCCATGCGGAACCTGGGAGAT CTGAGACTGGCCATTTAGGGGAGGGG	6618
		CCCTCCCC <u>T</u> AAATGGCC	6619
		GGCCATTT <u>A</u> GGGGAGGG	6620
	Increased stearate stearoyl-ACP desaturase	GCTCTGAGACTGAACCCTATCCCCACCCAAACCTTCTCCCTCC	6621
10	Glycine max Arg22Term AGA-TGA	TATTTTCAACCTCTTTGGAACCGGAGCGGAGGGTAGAGGCCATGC GGAACCTGGGAGATCAGACTGGCCATTTGGGGGAGGGAGAAG GTTTGGGTGGGGATAGGGTTCAGTCTCAGAGC	6622
		CCAGTCTCTGATCTCCC	6623
		GGGAGATC <u>A</u> GAGACTGG	6624
15	Increased stearate stearoyl-ACP desaturase	CAAATGGCCAGTCTCAGATCTCCCAGGTTCCGCATGGCCTCTACC CTCCGCTCCG	6625
	Glycine max Lys37Term AAA-TAA	TTACTTGAACATGCACTTCTCTGGGAGGAGTGAATGGCTTCTTAAT ATTTTCAACCTCTT A GGAACCGGAGCGAGGGTAGAGGCCATGCG GAACCTGGGAGATGTGAGACTGGCCATTTG	6626
		CCGGTTCC <u>T</u> AAGAGGTT	6627
		AACCTCTT <u>A</u> GGAACCGG	6628
20	Increased stearate stearoyl-ACP desaturase	CAACAAGCACACAAGAACAACATCAACAATGGCGATTCGCATCA ATACGGCGACGTTTTAATCAGACCTGTACCGTTCATTCGCGTTTCC TCAACCGAAACCTCTCAGATCTCCCAAAT	6629
	Helianthus annuus Gln11Term CAA-TAA	ATTTGGGAGATCTGAGAGGTTTCGGTTGAGGAAACGCGAATGAAC GGTACAGGTCTGATT <u>A</u> AAACGTCGCCGTATTGATGCGAATCGCCA TTGTTGATGTTGTTCTTGTGTGTGTGTTG	6630
		CGACGTTT <u>T</u> AATCAGAC	6631
		GTCTGATT <u>A</u> AAACGTCG	6632
25	Increased stearate stearoyl-ACP desaturase	AAGCACACAAGAACAACATCAACAATGGCGATTCGCATCAATAC GGCGACGTTTCAATGAGACCTGTACCGTTCATTCGCGTTTCCTCAA CCGAAACCTCTCAGATCTCCCAAATTCGC	6633
30	Helianthus annuus Ser12Term TCA-TGA	GCGAATTTGGGAGATCTGAGAGGTTTCGGTTGAGGAAACGCGAAT GAACGGTACAGGTCTCATTGAAACGTCGCCGTATTGATGCGAATC GCCATTGTTGATGTTGTTGTGTGTGCTT	6634

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	GTTTCAAT G AGACCTGT	6635
	ACAGGTCTCATTGAAAC	6636
ncreased stearate stearoyl-ACP	AAGAACAACATCAACAATGGCGATTCGCATCAATACGGCGACGTTT CAATCAGACCTGTAGCGTTCATTCGCGTTTCCTCAACCGAAACCTC TCAGATCTCCCAAATTCGCCATGGCTTCC	6637
desaturase Helianthus annuus Tyr15Term	GGAAGCCATGGCGAATTTGGCGTGGGGGGGGGTTTCGGTTGAGGGGAACCATGGCGAATTTGGGAGATCTGATGAAACGTCGCCGTATTGATGCGAATCGCCATTGTTGATGTTCTT	6638
TAC-TAG	GACCTGTAGCGTTCATT	6639
	AATGAACGCTACAGGTC	6640
Increased stearate stearoyl-ACP	CAACATCAACAATGGCGATTCGCATCAATACGGCGACGTTTCAATC AGACCTGTACCGTTGATTCGCGTTTCCTCAACCGAAACCTCTCAGA TCTCCCAAATTCGCCATGGCTTCCACCAT	6641
desaturase Helianthus annuus Ser17Term	ATGGTGGAAGCCATGGCTTCGACGAT ATGGTGGAAGCCATGGCGAATTTGGGAGATCTGAGAGGTTTCGGT TGAGGAAACGCGAATCAACGGTACAGGTCTGATTGAAACGTCGCC GTATTGATGCGAATCGCCATTGTTGATGTTG	6642
TCA-TGA	GTACCGTT <u>G</u> ATTCGCGT	6643
	ACGCGAATCAACGGTAC	6644
Increased stearate stearoyl-ACP	ACACACACACACACTCAATCACACACACATCATCATCTTCT	6645
desaturase Helianthus annuus Arg4Term	TCGATTGAAAAGTGTATGAAGGATATATCTCCCGTTGAAGCGT CACCGGACTCATTCAAAGCGCCATCGTTGATGAAGAAGATGATGA TGTGTGTGTGATTGAGTGTGTGT	6646
CGA-TGA	TGGCGCTTTGAATGAGT	6647
	ACTCATTCAAAGCGCCA	664
Increased stearate stearoyl-ACP	ACACACACATCATCATCTTCTTCATCAACGATGGCGCTTCGAATGA GTCCGGTGACGCTTTAACGGGAGATATATCCTTCATACACTTTTCA TCAATCGAAAAATCTCAGATCTCCTAAAT	
desaturase Helianthus annuus Gln11Term	ATTTAGGAGATCTGAGATTTTCGATTGAAAAGTGTATGAAGG ATATATCTCCCGTTAAAGCGTCACCGGACTCATTCGAAGCGCCAT CGTTGATGAAGAAGATGATGATGTGTGTGT	665
CAA-TAA	TGACGCTT <u>T</u> AACGGGAG	665
	CTCCCGTT A AAGCGTCA	665

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	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQID NO:
	Increased stearate stearoyl-ACP desaturase	ACATCATCTTCTTCATCAACGATGGCGCTTCGAATGAGTCCGG TGACGCTTCAACGGTAGATATATCCTTCATACACTTTTCATCAATCG AAAAATCTCAGATCTCCTAAATTCGCGA	6653
5	Helianthus annuus Glu13Term GAG-TAG	TCGCGAATTTAGGAGATCTGAGATTTTTCGATTGATGAAAAGTGTA TGAAGGATATATCT <u>A</u> CCGTTGAAGCGTCACCGGACTCATTCGAAG CGCCATCGTTGATGAAGAAGATGATGT	6654
		TTCAACGGTAGATATAT	6655
		ATATATCT <u>A</u> CCGTTGAA	6656
	Increased stearate stearoyl-ACP desaturase	ATCTTCTTCATCAACGATGGCGCTTCGAATGAGTCCGGTGACGCTT CAACGGGAGATATAGCCTTCATACACTTTTCATCAATCGAAAAATCT CAGATCTCCTAAATTCGCGATGGCTTCC	6657
10	Helianthus annuus Tyr15Term TAT-TAG	GGAAGCCATCGCGAATTTAGGAGATCTGAGATTTTTCGATTGATGA AAAGTGTATGAAGGCTATATCTCCCGTTGAAGCGTCACCGGACTC ATTCGAAGCGCCATCGTTGATGAAGAAGAT	6658
		GAGATATA <u>G</u> CCTTCATA	6659
		TATGAAGG <u>C</u> TATATCTC	6660
15	Increased stearate stearoyl-ACP desaturase	ACTCAGCCAGCTTGCCCCCAAACAACAGCGCAGAAAAACCTTCA ACAACAATGGCTCTCTAGCTCAACCCAGTCACCACCTTCCCTTCAA CACGCTCCCTCAACAACTTCTCCTCCAGAT	6661
	Linum usitatissimum Lys4Term AAG-TAG	ATCTGGAGGAGAGTTGTTGAGGGAGCGTGTTGAAGGGAAGGTG GTGACTGGGTTGAGCTAGAGGCCATTGTTGTTGAAGGTTTTTCTG CGCTGTTGTTTGGGGGCCAAGCTGGCTGAGTT	6662
		TGGCTCTCTAGCTCAAC	6663
		GTTGAGCT <u>A</u> GAGAGCCA	6664
20	Increased stearate stearoyl-ACP desaturase	GCGCAGAAAACCTTCAACAACAATGGCTCTCAAGCTCAACCCAG TCACCACCTTCCCTT G AACACGCTCCCTCAACAACTTCTCCTCCAG ATCTCCTCGCACCTTTCTCATGGCTGCTTC	6665
	Linum usitatissimum Ser13Term TCA-TGA	GAAGCAGCCATGAGAAAGGTGCGAGGAGATCTGGAGGAGAAGTT GTTGAGGGAGCGTGTT <u>C</u> AAGGGAAGGTGGTGACTGGGTTGAGCT TGAGAGCCATTGTTGTTGAAGGTTTTTCTGCGC	6666
		CTTCCCTT <u>G</u> AACACGCT	6667
		AGCGTGTT <u>C</u> AAGGGAAG	6668
25	Increased stearate stearoyl-ACP desaturase	CTCAAGCTCAACCCAGTCACCACCTTCCCTTCAACACGCTCCCTC AACAACTTCTCCTCCTGATCTCCTCGCACCTTTCTCATGGCTGCTT CCACTTTCAATTCCACCTCCACCAAGTAAG	6669
30	Linum usitatissimum Arg23Term AGA-TGA	CTTACTTGGTGGAGGTGGAATTGAAAGTGGAAGCAGCCATGAGAA AGGTGCGAGGAGATCAGGAGGAGGAGGTTGTTGAGGGAGCGTGTT GAAGGGAAGG	6670

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	TCTCCTCC <u>T</u> GATCTCCT	6671
	AGGAGATC <u>A</u> GGAGGAGA	6672
	TCCTCCAGATCTCCTCGCACCTTTCTCATGGCTGCTTCCACTTTCA	6673
ncreased stearate	ATTCCACCTCCACCTAGTAAGCATCTCCTCCTCCTCGGAATCTCCG	
tearoyl-ACP esaturase	ICCGATTTCTTTAAGCGATTGATCGTAGA	
inum usitatissimum	TCTACGATCAATCGCTTAAAAGAAATCGGCGGAGATTCCGAGGAG	6674
ys41Term	GAGGAGATGCTTACTAGGTGGAGGTGGAATTGAAAGTGGAAGCAG	
AG-TAG	CCATGAGAAAGGTGCGAGGAGATCTGGAGGA	6675
	CCTCCACCTAGTAAGCA	
	TGCTTACT <u>A</u> GGTGGAGG	6676
ncreased stearate	ATGGCACTGAAACTTTGCTTTCCACCCCACAAGATGCCTTCCTT	6677
stearoyl-ACP	CCGATGCTCGTATCTGATCTCACAGGGTTTTCATGGCTTCAACTAT	
desaturase	TCATTCTCCTTCTATGGAGGTCGGAAAAG	
Olea europaea	CTTTTCCGACCTCCATAGAAGGAGAATGAATAGTTGAAGCCATGAA	6678
Arg21Term	AACCCTGTGAGATCAGATACGAGCATCGGGGAAGGAAGGCATCTT	
AĞA-TGA	GTGGGGTGGAAAGCAAAGTTTCAGTGCCAT	6679
	CTCGTATC <u>T</u> GATCTCAC	
	GTGAGATC <u>A</u> GATACGAG	6680
Increased stearate	CCCACAAGATGCCTTCCTTCCCCGATGCTCGTATCAGATCTCACA	6681
stearoyl-ACP	GGGTTTTCATGGCTTGAACTATTCATTCTCCTTCTATGGAGGTCGG	
desaturase	AAAAGTTAAAAAGCCTTTCACGCCTCCACG	6682
Olea europaea	CGTGGAGGCGTGAAAGGCTTTTTAACTTTTCCGACCTCCATAGAAG	0004
Ser29Term	GAGAATGAATAGTTCAAGCCATGAAAACCCTGTGAGATCTGATACG	
TCA-TGA	AGCATCGGGGAAGGAAGGCATCTTGTGGG	668
	CATGGCTT <u>G</u> AACCCATC	668
	GAATAGTT <u>C</u> AAGCCATG	668
Increased stearate	GATGCTCGTATCAGATCTCACAGGGTTTTCATGGCTTCAACTATTC ATTCTCCTTCTATGTAGGTCGGAAAAGTTAAAAAGCCTTTCACGCC	000
stearoyl-ACP	ATTCTCCTTCTATGTAGGTCGGAAAAGTTAAAAAGGGTTTGAGGGG	
desaturase	TCCACGAGAGGTACATGTTCAAGTAACCC GGGTTACTTGAACATGTACCTCTCGTGGAGGCGTGAAAGGCTTTTT	668
Olea europaea	AACTTTTCCGACCTACATAGAAGGAGAATGAATAGTTGAAGCCATG	
Glu37Term GAG-TAG	AAAACCCTGTGAGATCTGATACGAGCATC	
IGAG-TAG	CTTCTATG <u>T</u> AGGTCGGA	668
	TCCGACCTACATAGAAG	668

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	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	Increased stearate stearoyl-ACP desaturase	CGTATCAGATCTCACAGGGTTTTCATGGCTTCAACTATTCATTC	6689
5	Olea europaea Gly39Term GGA-TGA	AGGAATGGGTTACTTGAACATGTACCTCTCGTGGAGGCGTGAAAG GCTTTTTAACTTTTCAGACCTCCATAGAAGGAGAATGAAT	6690
		TGGAGGTC <u>T</u> GAAAAGTT	6691
		AACTTTTC <u>A</u> GACCTCCA	6692
	Increased stearate stearoyl-ACP desaturase	TTCTCGTTTTTGTCGTCCCCTCTGCTCTCTCTCTCTATCAGGCACG GAGAAATGGCACTGTAACTCAGTCCAGTC	6693
10	Persea americana Lys4Term AAA-TAA	AAGGCGGATAGGAGGCAAGAAATGGAAGCTTCTGAGATTGAAACA TGACTGGACTG	6694
		TGGCACTG <u>T</u> AACTCAGT	6695
		ACTGAGTT <u>A</u> CAGTGCCA	6696
15	Increased stearate stearoyl-ACP desaturase	CTGCTCTCTCTCTATCAGGCACGGAGAAATGGCACTGAAACTC AGTCCAGTCATGTTTTAATCTCAGAAGCTTCCATTTCTTGCCTCCTA TCCGCCTTCCAATCTCAGATCTCCGAGGG	6697
	Persea americana Gln11Term CAA-TAA	CCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGGCAAGAAAT GGAAGCTTCTGAGATTAAAACATGACTGGACTG	6698
		TCATGTTT <u>T</u> AATCTCAG	6699
		CTGAGATT <u>A</u> AAACATGA	6700
20	Increased stearate stearoyl-ACP desaturase	TCTCTCTATCAGGCACGGAGAAATGGCACTGAAACTCAGTCCA GTCATGTTTCAATCTTAGAAGCTTCCATTTCTTGCCTCCTATCCGC CTTCCAATCTCAGATCTCCGAGGGTTTTCA	6701
	Persea americana Gln13Term CAG-TAG	TGAAAACCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGGCAA GAAATGGAAGCTTCT <u>A</u> AGATTGAAACATGACTGGACTGAGTTTCAG TGCCATTTCTCCGTGCCTGATAGAGAGAGA	6702
		TTCAATCT <u>T</u> AGAAGCTT	6703
		AAGCTTCT A AGATTGAA	6704
25	Increased stearate stearoyl-ACP desaturase	CTCTCTATCAGGCACGGAGAAATGGCACTGAAACTCAGTCCAGTC ATGTTTCAATCTCAGTAGCTTCCATTTCTTGCCTCCTATCCGCCTTC CAATCTCAGATCTCCGAGGGTTTTCATGG	6705
30	Persea americana Lys14Term AAG-TAG	CCATGAAAACCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGG CAAGAAATGGAAGCT <u>A</u> CTGAGATTGAAACATGACTGGACTGAGTTT CAGTGCCATTTCTCCGTGCCTGATAGAGAG	6706

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	AATCTCAG <u>T</u> AGCTTCCA	6707
	TGGAAGCT <u>A</u> CTGAGATT	6708
	CCCGAGATCTCGCTGCCGCTGCTCATGGCGTTCGCGGCGTCCC	6709
ncreased stearate tearoyl-ACP	LACACCGCATCGCCGTAGTCCTGCGGCGCGTGGCGCAGAGGAG	
lesaturase O <i>ryza sativa</i> Fyr12Term	GAGCAATGGGATGTCGAAGATGGTGGCCATGGCC GGCCATGGCCACCATCTTCGACATCCCATTGCTCCTCTCTGCGC CACGCCGCCGCAGGACTACGGCGATGCGGGGACGCCGCG	6710
TAC-TAG	AACGCCATGAGCAGCGGCAGCGAGATCTCGGGG TCGCCGTAGTCCTGCGG	6711
	CCGCAGGACTACGGCGA	6712
ncreased stearate stearoyl-ACP	CTGCTCATGGCGTTCGCGGCGTCCCACACCGCATCGCCGTACTC CTGCGGCGCGCGTGGCGTAGAGGAGGAGCAATGGGATGTCGAAGA	6713
desaturase O <i>ryza sativa</i> Gln19Term	TGGTGGCCATGGCCTCCACCATCAACAGGGTCA TGACCCTGTTGATGGTGGAGGCCATGGCCACCATCTTCGACATCC CATTGCTCCTCTACGCCACGCC	6714
CAG-TAG	GCGTGTGGGACGCCCCCTTGCCCTTGCCCTTGCCCTTGCGCGTAGAGGAGG	6715
	CCTCCTCTACGCCACGC	6716
Increased stearate stearoyl-ACP	CCCACACCGCATCGCCGTACTCCTGCGGCGCGTGGCGCAGAG GAGGAGCAATGGGATGTAGAAGATGGTGGCCATGGCCTCCACCA TCAACAGGGTCAAGACTGCTAAGAAGCCCTACAC	6717
desaturase Oryza sativa Ser26Term	GTGTAGGGTCAAGACTGCTAAGACTGCTAGACTGTGATGGTGGAGGCC GTGTAGGGCTTCTTAGCAGTCTTGACCCTGTTGATGGTGGAGGCC ATGGCCACCATCTTCTACATCCCATTGCTCCTCCTCTGCGCCACG CCGCCGCAGGAGTACGGCGATGCGGTGTGGG	6718
TCG-TAG	TGGGATGTAGAAGATGG	6719
	CCATCTTCTACATCCCA	672
Increased stearate stearoyl-ACP	CACACCGCATCGCCGTACTCCTGCGGCGCGTGGCGCAGAGGAG GAGCAATGGGATGTCGTAGATGGTGGCCATGGCCTCCACCATCAA CAGGGTCAAGACTGCTAAGAAGCCCTACACTC	
desaturase Oryza sativa Lys27Term	GAGGGTCAAGACTOOTTAGACTOTTGACCCTGTTGATGGTGGAGG GAGTGTAGGGCTTCTTAGCAGTCTTGACCCTGTTGATGGTGGAGG CCATGGCCACCATCTACGACATCCCATTGCTCCTCTGCGCCA CGCCGCCGCAGGAGTACGGCGATGCGGTGTG	
AAG-TAG	GGATGTCGTAGATGGTG	672
	CACCATCTACGACATCC	672

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SE N
Increased stearate stearoyl-ACP desaturase	TTCTCTCTAGGTTGAGCGGTTACCAACAGAAGCACTTAGGAGA GAGAAGCAATGGCGT <u>A</u> GAAGCTTCACCACACGGCCTTCAATCCTT CCATGGCGGTTACCTCTTCGGGACTTCCTCG	67
Simmondsia chinensis Leu3Term TTG-TAG	CGAGGAAGTCCCGAAGAGGTAACCGCCATGGAAGGATTGAAGGC CGTGTGGTGAAGCTTC <u>T</u> ACGCCATTGCTTCTCTCCTAAGTGCTT CTGTTGGTAACCGCTCAACCTAGAGAGAGAA	67
	AATGGCGT <u>A</u> GAAGCTTC	67
	GAAGCTTC <u>T</u> ACGCCATT	67
Increased stearate stearoyl-ACP desaturase	CTCTCTCTAGGTTGAGCGGTTACCAACAGAAGCACTTAGGAGAGA GAAGCAATGGCGTTGTAGCTTCACCACACGGCCTTCAATCCTTCC ATGGCGGTTACCTCTTCGGGACTTCCTCGAT	67
Simmondsia chinensis Lys4Term AAG-TAG	ATCGAGGAAGTCCCGAAGAGGTAACCGCCATGGAAGGATTGAAGGCCGTGTGGTGAAGCTACAACGCCATTGCTTCTCTCCTAAGTGCTTCTGTTGGTAACCGCTCAACCTAGAGAGAG	67
	TGGCGTTG <u>T</u> AGCTTCAC	67
	GTGAAGCT <u>A</u> CAACGCCA	67
Increased stearate stearoyl-ACP desaturase	AAGCAATGGCGTTGAAGCTTCACCACACGGCCTTCAATCCTTCCAT GGCGGTTACCTCTTAGGGACTTCCTCGATCGTATCACCTCAGATC TCACCGCGTTTTCATGGCTTCTTCTACAAT	67
Simmondsia chinensis Ser19Term TCG-TAG	ATTGTAGAAGAAGCCATGAAAACGCGGTGAGATCTGAGGTGATAC GATCGAGGAAGTCCC <u>T</u> AAGAGGTAACCGCCATGGAAGGATTGAAG GCCGTGTGGTGAAGCTTCAACGCCATTGCTT	67
	TACCTCTTAGGGACTTC	67
	GAAGTCCC <u>T</u> AAGAGGTA	67
Increased stearate stearoyl-ACP desaturase	GCAATGGCGTTGAAGCTTCACCACACGGCCTTCAATCCTTCCATG GCGGTTACCTCTTCGTGACTTCCTCGATCGTATCACCTCAGATCTC ACCGCGTTTTCATGGCTTCTTCTACAATTG	67
Simmondsia chinensis Gly20Term GGA-TGA	CAATTGTAGAAGAAGCCATGAAAACGCGGTGAGATCTGAGGTGAT ACGATCGAGGAAGTC <u>A</u> CGAAGAGGTAACCGCCATGGAAGGATTGA AGGCCGTGTGGTGAAGCTTCAACGCCATTGC	67
	CCTCTTCGTGACTTCCT	67
	AGGAAGTC <u>A</u> CGAAGAGG	67
Increased stearate stearoyl-ACP desaturase	TGGCTCTGAATCTCAACCCCGTTTCCACACCATTTCAGTGTCGTCG ATTGCCGTCTTTCTGACCTCGTCAAACGCCTTCTCGCAGATCTCCC AAATTCTTCATGGCTTCCACTCTCAGCAG	67
Spinacia oleracea Ser21Term TCA-TGA	CTGCTGAGAGTGGAAGCCATGAAGAATTTGGGAGATCTGCGAGAA GGCGTTTGACGAGGT <u>C</u> AGAAAGACGGCAATCGACGACACTGAAAT GGTGTGGAAACGGGGTTGAGATTCAGAGCCA	67

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Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
Alteration	GTCTTTCT G ACCTCGTC	6743
	GACGAGGT <u>C</u> AGAAAGAC	6744
ncreased stearate tearoyl-ACP	AATCTCAACCCCGTTTCCACACCATTTCAGTGTCGTCGATTGCCGT CTTTCTCACCTCGTTAAACGCCTTCTCGCAGATCTCCCAAATTCTT	6745
lesaturase Spinacia oleracea Gln24Term	CATGGCTTCCACTCTCAGCAGCTCTTCTC GAGAAGAGCTGCTGAGAGTGGAAGCCATGAAGAATTTGGGAGATC TGCGAGAAGGCGTTTAACGAGGTGAGAAAGACGGCAATCGACGA CACTGAAATGGTGTGGAAACGGGGTTGAGATT	6746
CAA-TAA	CACTGAAATGGTGTGGAAACGCGGTTG, IGACGCGTTGAAACGCCT	6747
	AGGCGTTT A ACGAGGTG	6748
Increased stearate stearoyl-ACP	TCCACACCATTTCAGTGTCGTCGATTGCCGTCTTTCTCACCTCGTC AAACGCCTTCTCCCTAAGGAAGCGGAAA	6749
desaturase <i>Spinacia oleracea</i> Arg29Term	TTTCCGCTTCCTTAGGAAGAGCCGTTT TTTCCGCTTCCTTAGGAGAGAGCCGTGAGAGCCATGA AGAATTTGGGAGATCAGCGAGAAGGCGTTTGACGAGGTGAGAAAG ACGCCAATCGACGACACTGAAATGGTGTGGA	6750
AGA-TGA	CTTCTCGCTGATCTCCC	6751
	GGGAGATC A GCGAGAAG	6752
Increased stearate stearoyl-ACP	TTTCAGTGTCGATTGCCGTCTTTCTCACCTCGTCAAACGCCTT CTCGCAGATCTCCCTAATTCTTCATGGCTTCCACTCTCAGCAGCTC TTCTCCTAAGGAAGCGGAAAGCCTGAAGA	6753
desaturase Spinacia oleracea Lys32Term AAA-TAA	TCTCCTAAGGAAGCGGAAAGCGTGAGAGAGAGCTGCTGAGAGTGG TCTTCAGGCTTTCCGCTTCCTTAGGAGAAGAGCTGCTGAGAGTGG AAGCCATGAAGAATTAGGGAGAGATCTGCGAGAAGGCGTTTGACGAG GTGAGAAAGACGGCAATCGACGACACTGAAA	6754
IAAA-IAA	GATCTCCC <u>T</u> AATTCTTC	675
	GAAGAATT <u>A</u> GGGAGATC	675
Increased stearate stearoyl-ACP	AAATAGTCGAGGTGAAAAACAGAGCATCAACAATGGCACTGAATAT CAATGGGGTGTCGTGAAAATCTCACAAAATGTTACCATTTCCTTGT TCTTCAGCCAGATCTGAGCGAGTTTTCAT	675
desaturase Solanum tuberosum Leu10Term	ATGAAAACTCGCTCAGATCTGGCTGAAGAACAAGGAAATGGTAACA TTTTGTGAGATTTTCACGACACCCCATTGATATTCAGTGCCATTGTT	675
TTA-TGA	GATGCTCTGTTTTCACCTCGACTATTT GGTGTCGTGAAAATCTC	675
	GAGATTTT <u>C</u> ACGACACC	676

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ I NO:
Increased stearate stearoyl-ACP desaturase	ATAGTCGAGGTGAAAAACAGAGCATCAACAATGGCACTGAATATCA ATGGGGTGTCGTTA <u>T</u> AATCTCACAAAATGTTACCATTTCCTTGTTCT TCAGCCAGATCTGAGCGAGTTTTCATGG	676′
Solanum tuberosum Lys11Term AAA-TAA	CCATGAAAACTCGCTCAGATCTGGCTGAAGAACAAGGAAATGGTA ACATTTTGTGAGATTATAACGACACCCCATTGATATTCAGTGCCATT GTTGATGCTCTGTTTTTCACCTCGACTAT	676
	TGTCGTTATAATCTCAC	676
	GTGAGATT <u>A</u> TAACGACA	676
Increased stearate stearoyl-ACP desaturase	GTGAAAAACAGAGCATCAACAATGGCACTGAATATCAATGGGGTGT CGTTAAAATCTCAC <u>T</u> AAATGTTACCATTTCCTTGTTCTTCAGCCAGA TCTGAGCGAGTTTTCATGGCTTCAACCA	676
Solanum tuberosum Lys14Term AAA-TAA	TGGTTGAAGCCATGAAAACTCGCTCAGATCTGGCTGAAGAACAAG GAAATGGTAACATTTAGTGAGATTTTAACGACACCCCATTGATATTC AGTGCCATTGTTGATGCTCTGTTTTTCAC	676
	AATCTCAC <u>T</u> AAATGTTA	676
	TAACATTT <u>A</u> GTGAGATT	676
Increased stearate stearoyl-ACP desaturase	ACAGAGCATCAACAATGGCACTGAATATCAATGGGGTGTCGTTAAA ATCTCACAAAATGT <u>G</u> ACCATTTCCTTGTTCTTCAGCCAGATCTGAG CGAGTTTTCATGGCTTCAACCATTCATCG	676
Solanum tuberosum Leu16Term TTA-TGA	CGATGAATGGTTGAAGCCATGAAAACTCGCTCAGATCTGGCTGAA GAACAAGGAAATGGT <u>C</u> ACATTTTGTGAGATTTTAACGACACCCCAT TGATATTCAGTGCCATTGTTGATGCTCTGT	677
	CAAAATGT <u>G</u> ACCATTTC	677
	GAAATGGT <u>C</u> ACATTTTG	677
Increased stearate stearoyl-ACP desaturase	TGGCTCTGAGGCTGAACCCTAACCCTTCACAGAAGCTCTTTCTCTC TCCTTCTTCATCATCATCGATCTTCTTCTTCTTCATCGTTCTCGCTTCCTC AAATGGCTAGCCTCAGATCTCCAAGGTT	677
<i>Arachis hypogaea</i> Ser21Term TCA-TGA	AACCTTGGAGATCTGAGGCTAGCCATTTGAGGAAGCGAGAACGAT GAAGAAGAAGAAGAT <u>C</u> ATGATGAAGAAGAGAGAGAGAAAGAGCTTC TGTGAAGGGTTAGGGTTCAGCCTCAGAGCCA	677
	TTCATCAT <u>G</u> ATCTTCTT	677
	AAGAAGAT <u>C</u> ATGATGAA	677
Increased stearate stearoyl-ACP desaturase	ACCCTAACCCTTCACAGAAGCTCTTTCTCTCTCCTTCTTCATCATCA TCTTCTTCTTCTTGATCGTTCTCGCTTCCTCAAATGGCTAGCCTCA GATCTCCAAGGTTCCGCATGGCCTCCAC	677
Arachis hypogaea Ser26Term TCA-TGA	GTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCTAGCCATTTGA GGAAGCGAGAACGATCAAGAAGAAGAAGATGATGAAGAAGGA GAGAGAAAGAGCTTCTGTGAAGGGTTAGGGT	677

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	TTCTTCTT G ATCGTTCT	6779
•		6780
ncreased stearate stearoyl-ACP	AGAACGATCAAGAAGAA CTAACCCTTCACAGAAGCTCTTTCTCTCTCTCTCATCATCATCT TCTTCTTCTTCATAGTTCTCGCTTCCTCAAATGGCTAGCCTCAGAT	6781
lesaturase A <i>rachis hypogaea</i> Ser27Term	CTCCAAGGTTCCGCATGGCCTCCACCCT AGGGTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCTAGCCAT TTGAGGAAGCGAGAACTATGAAGAAGAAGAAGATGATGATGAAGA AGGAGAGAAAGAGCTTCTGTGAAGGGTTAG	6782
rcg-tag	TTCTTCATAGTTCTCGC	6783
	GCGAGAAC <u>T</u> ATGAAGAA	6784
Increased stearate stearoyl-ACP	CTTCACAGAAGCTCTTCTCTCTCTCTTCATCATCATCTTCTTCT TCTTCATCGTTCTAGCTTCCTCAAATGGCTAGCCTCAGATCTCCAA	6785
desaturase Arachis hypogaea Ser29Term	GTTCCGCATGGCCTCCACCOTTCGCACCTTCGAGGATCTGAGGCT GTGCGGAGGGTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCT AGCCATTTGAGGAAGCTAGAACGATGAAGAAGAAGAAGATGATGA TGAAGAAGGAGAGAAAGAGCTTCTGTGAAG	6786
TCG-TAG	ATCGTTCTAGCTTCCTC	6787
i	GAGGAAGC <u>T</u> AGAACGAT	6788
Increased stearate stearoyl-ACP	AAAGTTAAAAGCCGTCCAAAACCCAAACCAGGAAAGGCAAACGAA AAGAAAAATGGCTT A GAATTTTAATGCCATCGCCTCGAAATCTCA	6789
desaturase Gossypium hirsutum Leu3Term	GAAGCTCCCTTGCTTTGCTCTTCCACCAAA TITGGTGGAAGAGCAAAGCAAGGGAGCTTCTGAGATTTCGAGGCG ATGGCATTAAAATTCTAAGCCATTTTTTCTTTTC	6790
TTG-TAG	AATGGCTT <u>A</u> GAATTITA	6791
	TAAAATTC <u>T</u> AAGCCATT	6792
Increased stearate stearoyl-ACP	CCCAAACCAGGAAAGGCAAACGAAAAGAAAAATGGCTTTGAATTT TAATGCCATCGCCTAGAAATCTCCGAA	679
desaturase Gossypium hirsutum Ser1-Term TCG-TAG		
	CATCGCCTAGAAATCTC	679
	GAGATTTC <u>T</u> AGGCGATG	679

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Increased stearate stearoyl-ACP desaturase	CAAACCAGGAAAGGCAAACGAAAAGAAAAATGGCTTTGAATTTTA ATGCCATCGCCTCGTAATCTCAGAAGCTCCCTTGCTTTGCTCTTCC ACCAAAGGCCACCCTTAGATCTCCCAAGT	6797
Gossypium hirsutum Lys11Term AAA-TAA	ACTTGGGAGATCTAAGGGTGGCCTTTGGTGGAAGAGCAAGCA	6798
	TCGCCTCG <u>T</u> AATCTCAG	6799
	CTGAGATT <u>A</u> CGAGGCGA	6800
Increased stearate stearoyl-ACP desaturase	AGGAAAGGCAAACGAAAAGAAAAAATGGCTTTGAATTTTAATGCCA TCGCCTCGAAATCT <u>T</u> AGAAGCTCCCTTGCTTTGCTCTTCCACCAAA GGCCACCCTTAGATCTCCCAAGTTTTCCA	6801
Gossypium hirsutum Gln13Term CAG-TAG	TGGAAAACTTGGGAGATCTAAGGGTGGCCTTTGGTGGAAGAGCAA AGCAAGGGAGCTTCT A AGATTTCGAGGCGATGGCATTAAAATTCAA AGCCATTTTTTCTTTTC	6802
	CGAAATCT <u>T</u> AGAAGCTC	6803
	GAGCTTCT <u>A</u> AGATTTCG	6804

Table 24
Oligonucleotides to produce plants with reduced linolenic acid

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ I NO:
omega-3 fatty acid	AATAGAACGACAGAGACTTTTCCTCTTTTCTTCTTGGGAAGAGGC TCCAATGGCGAGCTAGGTTTTATCAGAATGTGGTTTTAGACCTCTC CCCAGATTCTACCCTAAACACACACACCTC	6805
Arabidopsis thaliana Ser4Term	GAGGTTGTGTTTAGGGTAGAATCTGGGGAGAGGTCTAAAACCA CATTCTGATAAAACC <u>T</u> AGCTCGCCATTGGAGCCTCTTCCCAAGAAG AAAAGAGGAAAAAGTCTCTGTCGTTCTATT	680
TCG-TAG	GGCGAGCT <u>T</u> GGTTTTAT	680
1	ATAAAACC <u>A</u> AGCTCGCC	680
Reduced linolenic acid omega-3 fatty acid	ACGACAGAGACTTTTTCCTCTTTTCTTCTTGGGAAGAGGCTCCAAT GGCGAGCTCGGTTTGATCAGAATGTGGTTTTAGACCTCTCCCCAG	680
desaturase Arabidopsis thaliana Leu6Term	GCAAAAGAGGTTGTGTTTAGGGTAGAATCTGGGGAGAGGTCTA AAACCACATTCTGAT <u>C</u> AAACCGAGCTCGCCATTGGAGCCTCTTCC CAAGAAGAAAAAGGGAAAAAGTCTCTGTCGT	681
TTA-TGA	CTCGGTTTGATCAGAAT	68
	ATTCTGAT C AAACCGAG	68
Reduced linolenic acid omega-3 fatty acid	ACAGAGACTTTTTCCTCTTTTCTTCTTGGGAAGAGGCTCCAATGGC GAGCTCGGTTTTATGAGAATGTGGTTTTAGACCTCTCCCCAGATTC TACCCTAAACACACAACCTCTTTTGCCTC	68
desaturase Arabidopsis thaliana Ser7Term	GAGGCAAAAGAGGTTGTGTTTTAGGGTAGAATCTGGGGAGAGGT CTAAAACCACATTCTCATAAAACCGAGCTCGCCATTGGAGCCTCTT CCCAAGAAGAAAAGAGGAAAAAGTCTCTGT	68
TCA-TGA	GGTTTTAT <u>G</u> AGAATGTG	68
	CACATTCT C ATAAAACC	68
Reduced linolenic acid omega-3 fatty acid	GCTCGGTTTTATCATAATGTGGTTTTAGACCTCTCCCAGATTCTA	
desaturase Arabidopsis thaliana Glu8Term	TAGAGCAAAAGAGGTTGTGTTTAGGGTAGAATCTGGGGAGAG GTCTAAAACCACATTATGATAAAACCGAGCTCGCCATTGGAGCCTC TTCCCAAGAAGAAAAGGGAAAAAGTCTCT	<u> </u>
GAA-TAA	TTTTATCATAATGTGGT	6
	ACCACATTATGATAAAA	6

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID
	Reduced linolenic acid omega-3 fatty acid desaturase	TCATCATCTTCTTCTTCTGGGGAGAGAGAGAGAGCAAAAGAGCTCT AGCAATGGCGAACT <u>A</u> GGTCTTATCCGAATGTGGCATAAGACCTCT CCCCAGAATCTACACCACACC	6821
5	Brassica juncea Leu4Term TTG-TAG	GTGGATCTGGGTGTGGTGTAGATTCTGGGGAGAGGTCTTATGCCA CATTCGGATAAGACCTAGTTCGCCATTGCTAGAGCTCTTTTGCTCT CTCTCTCCCCCAGAAGAAGAAGATGATGA	6822
		GGCGAACT <u>A</u> GGTCTTAT	6823 6824
		ATAAGACC <u>T</u> AGTTCGCC	
	Reduced linolenic acid omega-3 fatty acid desaturase	TCTTCTTCTGGGGAGAGAGAGAGCAAAAGAGCTCTAGCAA TGGCGAACTTGGTCT <u>G</u> ATCCGAATGTGGCATAAGACCTCTCCCCA GAATCTACACCACACC	6825
10	Brassica juncea Leu6Term TTA-TGA	AGGAAAGTGGATCTGGGTGTGTGTAGATTCTGGGGAGAGGTCTT ATGCCACATTCGGAT <u>C</u> AGACCAAGTTCGCCATTGCTAGAGCTCTTT TGCTCTCTCTCTCCCCAGAAGAAGAAGA	6826
		CTTGGTCT <u>G</u> ATCCGAAT	6827
		ATTCGGAT <u>C</u> AGACCAAG	6828
15	Reduced linolenic acid omega-3 fatty acid desaturase	TTCTTCTGGGGAGAGAGAGAGAGCAAAAGAGCTCTAGCAATGGCG AACTTGGTCTTATCCTAATGTGGCATAAGACCTCTCCCCAGAATCT ACACCACACC	6829
	Brassica juncea Glu8Term GAA-TAA	TGGAGAGGAAAGTGGATCTGGGTGTGGTGTAGATTCTGGGGAGA GGTCTTATGCCACATTAGGATAAGACCAAGTTCGCCATTGCTAGAG CTCTTTTGCTCTCTCTCTCCCCAGAAGAA	6830
		TCTTATCC <u>T</u> AATGTGGC	6831
		GCCACATT <u>A</u> GGATAAGA	6832
20	Reduced linolenic acid omega-3 fatty acid desaturase	CTGGGGAGAGAGAGAGCAAAAGAGCTCTAGCAATGGCGAACTT GGTCTTATCCGAATGAGGCATAAGACCTCTCCCCAGAATCTACAC CACACCCAGATCCACTTTCCTCTCCAACACC	6833
	Brassica juncea Cys9Term TGT-TGA	GGTGTTGGAGAGGAAAGTGGATCTGGGTGTGGTGTAGATTCTGGG GAGAGGTCTTATGCCTCATTCGGATAAGACCAAGTTCGCCATTGCT AGAGCTCTTTTGCTCTCTCTCTCCCCAG	6834
		TCCGAATG <u>A</u> GGCATAAG	68 35
		CTTATGCC <u>T</u> CATTCGGA	6836
25	Reduced linolenic acid omega-3 fatty acid desaturase	ATAACAGAATTGCTGAATTCTTGCATTTTTAGCTTCTGGGTTTTCAA TGGCTGCTGGTTGAGTATTATCAGAATGTGGTTTAAGGCCTCTCCC AAGAATCTACTCACGACCCAGAATTGGT	6837
30	Ricinus communis Trp5Term TGG-TGA	ACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAGGCCTTAAACC ACATTCTGATAATAC <u>T</u> CAACCAGCAGCCATTGAAAACCCAGAAGCT AAAAATGCAAGAATTCAGCAATTCTGTTAT	6838

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Phenotype, Gene, Plant & Targeted	Altering Oligos	EQID NO:
Alteration	GCTGGTTG <u>A</u> GTATTATC	6839
	GATAATAC <u>T</u> CAACCAGC	6840
educed linolenic acid mega-3 fatty acid	AGAATTGCTGAATTCTTGCATTTTTAGCTTCTGGGTTTTCAATGGCT GCTGGTTGGGTAT G ATCAGAATGTGGTTTAAGGCCTCTCCCAAGA	6841
esaturase Ricinus communis eu7Term	ATCTACTCACGACCCAGAATTGGTTTTAC GTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAGGCCTT AAACCACATTCTGATCATACCCAACCAGCAGCCATTGAAAACCCAG AAGCTAAAAATGCAAGAATTCAGCAATTCT	6842
TA-TGA	TTGGGTATGATCAGAAT	6843
	ATTCTGATCATACCCAA	6844
Reduced linolenic acid omega-3 fatty acid	ATTGCTGAATTCTTGCATTTTTAGCTTCTGGGTTTTCAATGGCTGCT GGTTGGGTATTATGAGAATGTGGTTTAAGGCCTCTCCCAAGAATCT ACTCACGACCCAGAATTGGTTTTACATC	6845
desaturase Ricinus communis Ser8Term	GATGTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAGGC CTTAAACCACATTCTCATAATACCCAACCAGCAGCCATTGAAAACC CAGAAGCTAAAAATGCAAGAATTCAGCAAT	6846
TCA-TGA	GGTATTAT G AGAATGTG	6847
	CACATTCT <u>C</u> ATAATACC	6848
Reduced linolenic acid omega-3 fatty acid	TGCTGAATTCTTGCATTTTTAGCTTCTGGGTTTTCAATGGCTGCTG	6849
desaturase Ricinus communis Glu9Term	CTCACGACCCAGAATTGGTTTTACATCGA TCGATGTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAG GCCTTAAACCACATTATGATAATACCCAACCAGCAGCCATTGAAAA CCCAGAAGCTAAAAATGCAAGAATTCAGCA	6850
GAA-TAA	TATTATCA <u>T</u> AATGTGGT	685
	ACCACATT <u>A</u> TGATAATA	685
Reduced linolenic aci omega-3 fatty acid	TCTACCCTAAGCCCTGAACTGGGGGCAGCCACTTCTGCCTCTGTCTG	685
desaturase Nicotiana tabacum Arg22Term	GATCTGTACGTGAAATTCTCAACTTAATGTGAGAGGGGGGGAGAGT GGCTGCCCCAGTTCAGGGCTTAGGGTAGATTCTTGGGAGTGGTCT	685
AGA-TGA	AAGACCACATTCTGATAAAACCCAACTTGC CTAAGCCCTGAACTGGG	685
	CCCAGTTCAGGGCTTAG	685

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	Reduced linolenic acid omega-3 fatty acid desaturase	CTCCCAAGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCT GCCTCCTCTCACATTTAGTTGAGAATTTCACGTACAGATCTGAGTG GTTCTGCAATTTCTTTGTCTAATACTAATA	6857
5	Nicotiana tabacum Lys34Term AAG-TAG	TATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCTGTACG TGAAATTCTCAACTAAATGTGAGAGGGAGGCAGAAGTGGCTGCCCC AGTTCTGGGCTTAGGGTAGATTCTTGGGAG	6858
		CTCACATT <u>T</u> AGTTGAGA	6859
		TCTCAACT <u>A</u> AATGTGAG	6860
	Reduced linolenic acid omega-3 fatty acid desaturase	CAAGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCTGCCT CCTCTCACATTAAGTAGAGAATTTCACGTACAGATCTGAGTGGTTC TGCAATTTCTTTGTCTAATACTAATAAAGA	6861
10	Nicotiana tabacum Leu35Term TTG-TAG	TCTTTATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCTGT ACGTGAAATTCTC <u>T</u> ACTTAATGTGAGAGGGGCAGAAGTGGCTGC CCCAGTTCTGGGCTTAGGGTAGATTCTTG	6862
		CATTAAGT <u>A</u> GAGAATTT	6863
		AAATTCTC <u>T</u> ACTTAATG	6864
15	Reduced linolenic acid omega-3 fatty acid desaturase	AGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCTGCCTCC TCTCACATTAAGTTGTGAATTTCACGTACAGATCTGAGTGGTTCTG CAATTTCTTTGTCTAATACTAATAAAGAGA	6865
	Nicotiana tabacum Arg36Term AGA-TGA	TCTCTTTATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCT GTACGTGAAATTCACAACTTAATGTGAGAGGGGGCAGAAGTGGCT GCCCCAGTTCTGGGCTTAGGGTAGATTCT	6866
		TTAAGTTG <u>T</u> GAATTTCA	6867
		TGAAATTC <u>A</u> CAACTTAA	6868
20	Reduced linolenic acid omega-3 fatty acid desaturase	GCGAGTTGGGTTTTATCAGAATGTGGTCTGAGGCCACTCCCGAGG GTCTATCCTAAGCCA <u>T</u> GAACTGGCCACCCTTTGTTGAATTCCAATC CCACAAAGCTGAGATTTTCAAGAACAGATC	6869
	Sesamum indicum Arg22Term AGA-TGA	GATCTGTTCTTGAAAATCTCAGCTTTGTGGGATTGGAATTCAACAA AGGGTGGCCAGTTCATGGCTTAGGATAGACCCTCGGGAGTGGCC TCAGACCACATTCTGATAAAACCCAACTCGC	6870
		CTAAGCCA <u>T</u> GAACTGGC	6871
		GCCAGTTC <u>A</u> TGGCTTAG	6872
25	Reduced linolenic acid omega-3 fatty acid desaturase	CAGAATGTGGTCTGAGGCCACTCCCGAGGGTCTATCCTAAGCCAA GAACTGGCCACCCTT <u>A</u> GTTGAATTCCAATCCCACAAAGCTGAGATT TTCAAGAACAGATCTTGGAAATGGTTCTTC	6873
30	Sesamum indicum Leu27Term TTG-TAG	GAAGAACCATTTCCAAGATCTGTTCTTGAAAATCTCAGCTTTGTGG GATTGGAATTCAACTAAGGGTGGCCAGTTCTTGGCTTAGGATAGA CCCTCGGGAGTGGCCTCAGACCACATTCTG	6874
50	F-1-2-1/10	TOOLIANDAOAOAITOIG	<u>ئـــــــ</u>

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	CCACCCTT A GTTGAATT	6875
1	AATTCAAC <u>T</u> AAGGGTGG	6876
teduced linolenic acid mega-3 fatty acid	AATGTGGTCTGAGGCCACTCCCGAGGGTCTATCCTAAGCCAAGAA CTGGCCACCCTTTGTAGAATTCCAATCCCACAAAGCTGAGATTTTC AAGAACAGATCTTGGAAATGGTTCTTCATT	6877
esaturase Gesamum indicum eu28Term	AAGAACAGATCTTGGAAATCGTTGTTGAAAATCTCAGCTTTGT AATGAAGAACCATTTCCAAGATCTGTTCTTGAAAATCTCAGCTTTGT GGGATTGGAATTCTACAAAGGGTGGCCAGTTCTTGGCTTAGGATA GACCCTCGGGAGTGGCCTCAGACCACATT	6878
TG-TAG	CCCTTGTAGAATTCCA	6879
	TGGAATTC <u>T</u> ACAAAGGG	6880
Reduced linolenic acid omega-3 fatty acid	CTCCCGAGGGTCTATCCTAAGCCAAGAACTGGCCACCCTTTGTTG AATTCCAATCCCACATAGCTGAGATTTTCAAGAACAGATCTTGGAA ATGGTTCTTCATTCTGTTTGTCGAGTGGGA	6881
desaturase Sesamum indicum Lys34Term	TCCCACTCGACAAACAGAATGAAGAACCATTTCCAAGATCTGTTCT TGAAAATCTCAGCTATGTGGGATTGGAATTCAACAAAGGGTGGCC AGTTCTTGGCTTAGGATAGACCCTCGGGAG	6882
AAG-TAG	ATCCCACATAGCTGAGA	6883
	TCTCAGCTATGTGGGAT	6884
Reduced linolenic acid omega-3 fatty acid	CATCAGAGCGGCGATACCTAAGCATTGCTGGGTTAAGAATCCATG GAAGTCTATGAGTTAGGTCGTCAGAGAGCTAGCCATCGTGTTCGC ACTAGCTGCTGGAGCTGCTTACCTCAACAAT	-6885
desaturase Brassica napus Tyr3Term TAC-TAG	ATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGAACACGATGGC TAGCTCTCTGACGACCTAACTCATAGACTTCCATGGATTCTTAACC CAGCAATGCTTAGGTATCGCCGCTCTGATG	6886
TAC-TAG	ATGAGTTA <u>G</u> GTCGTCAG	688
	CTGACGAC <u>C</u> TAACTCAT	688
Reduced linolenic acid	ATGAGTTACGTCGTGTGAGAGCTAGCCATCGTGTTCGCACTAGCT	688
desaturase Brassica napus Arg6Term AGA-TGA	CAAGCCAATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGAACA CGATGGCTAGCTCTCAGACGACGTAACTCATAGACTTCCATGGAT TCTTAACCCAGCAATGCTTAGGTATCGCCGC	689
	ACGTCGTCTGAGAGCTA	689
	TAGCTCTCAGACGACGT	689

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID
Reduced linolenic acid omega-3 fatty acid desaturase	GCGATACCTAAGCATTGCTGGGTTAAGAATCCATGGAAGTCTATGA GTTACGTCGTCAGA <u>T</u> AGCTAGCCATCGTGTTCGCACTAGCTGCTG GAGCTGCTTACCTCAACAATTGGCTTGTTT	6893
<i>Brassica napus</i> Glu7Term GAG-TAG	AAACAAGCCAATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGA ACACGATGGCTAGCT <u>A</u> TCTGACGACGTAACTCATAGACTTCCATG GATTCTTAACCCAGCAATGCTTAGGTATCGC	6894
	TCGTCAGA <u>T</u> AGCTAGCC	6895
	GGCTAGCT <u>A</u> TCTGACGA	6896
Reduced linolenic acid omega-3 fatty acid desaturase	CCATGGAAGTCTATGAGTTACGTCGTCAGAGAGCTAGCCATCGTG TTCGCACTAGCTGCTTGAGCTGCTTACCTCAACAATTGGCTTGTTT GGCCTCTCTATTGGATTGCTCAAGGAACCA	6897
<i>Brassica napus</i> Gly17Term GGA-TGA	TGGTTCCTTGAGCAATCCAATAGAGAGGCCAAACAAGCCAATTGTT GAGGTAAGCAGCTCAAGCAGCTAGTGCGAACACGATGGCTAGCT CTCTGACGACGTAACTCATAGACTTCCATGG	6898
	TAGCTGCT <u>T</u> GAGCTGCT	6899
	AGCAGCTC <u>A</u> AGCAGCTA	6900
Reduced linolenic acid omega-3 fatty acid desaturase	GCAAGTTGGGTTCTATCAGAATGTGGTCTTAGACCACTACCAAGAA TATACCCAAAGCCC <u>T</u> GAATAGGGTCTTCTTCCGTTTGCGCCACCAA TTTAAATCTGAGAAGAATTTCACCTTCAC	6901
Solanum tuberosum Arg22Term AGA-TGA	GTGAAGGTGAAATTCTTCTCAGATTTAAATTGGTGGCGCAAACGGA AGAAGACCCTATTCAGGGCTTTGGGTATATTCTTGGTAGTGGTCTA AGACCACATTCTGATAGAACCCAACTTGC	6902
	CAAAGCCC <u>T</u> GAATAGGG	6903
	CCCTATTC <u>A</u> GGGCTTTG	6904
Reduced linolenic acid omega-3 fatty acid desaturase	TGGTCTTAGACCACTACCAAGAATATACCCAAAGCCCAGAATAGG GTCTTCTTCCGTTTGAGCCACCAATTTAAATCTGAGAAGAATTTCA CCTTCACCTATACGAACAGATCGGAATTGT	6905
Solanum tuberosum Cys29Term TGC-TGA	ACAATTCCGATCTGTTCGTATAGGTGAAGGTGAAATTCTTCTCAGA TTTAAATTGGTGGCTCAAACGGAAGAAGACCCTATTCTGGGCTTTG GGTATATTCTTGGTAGTGGTCTAAGACCA	6906
	TCCGTTTG <u>A</u> GCCACCAA	6907
	TTGGTGGC <u>T</u> CAAACGGA	6908
Reduced linolenic acid omega-3 fatty acid desaturase	CACTACCAAGAATATACCCAAAGCCCAGAATAGGGTCTTCTTCCGT TTGCGCCACCAATT G AAATCTGAGAAGAATTTCACCTTCACCTATA CGAACAGATCGGAATTGTTGGGCATTGAG	6909
Solanum tuberosum Leu33Term TTA-TGA	CTCAATGCCCAACAATTCCGATCTGTTCGTATAGGTGAAGGTGAAA TTCTTCTCAGATTTCAATTGGTGGCGCAAACGGAAGAAGACCCTAT TCTGGGCTTTGGGTATATTCTTGGTAGTG	6910

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Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQID NO:
Alteration	CACCAATT G AAATCTGA	6911
t	TCAGATIT <u>C</u> AATTGGTG	6912
	AGAATATACCCAAAGCCCAGAATAGGGTCTTCTTCCGTTTGCGCCA CCAATTTAAATCTGTGAAGAATTTCACCTTCACCTATACGAACAGAT	6913
esaturase olanum tuberosum rg36Term	CGGAATTGTTGGGCATTGAGGGTAAGTG CACTTACCCTCAATGCCCAACAATTCCGATCTGTTCGTATAGGTGA AGGTGAAATTCTTCACAGATTTAAATTGGTGGCGCAAACGGAAGAA	6914
GA-TGA	GACCCTATTCTGGGCTTTGGGTATATTCT TAAATCTGTGAAGAATT	6915
	AATTCTTC <u>A</u> CAGATTTA	6916
Reduced linolenic acid omega-3 fatty acid	CTCTTTATTATCCTCCTCTTCTTTGTTTTTTTGAGTTCTGAGTCACC TATGGCAAGTTGAGTGATTTCAGAATGTGGGCTAAGGCCACTTCC	6917
desaturase Petroselinum crispum Frp4Term	TCCACTTCTGGGCCTGGCATAGATTCTTGGAAGTGGCCTTAGCCC ACATTCTGAAATCACTCAACTTGCCATAGGTGACTCAGAACTCAAA AAAAACAAAGAAGAGGGGGGATAATAAAGAG	6918
rgg-tga	GCAAGTTGAGTGATTTC	6919
	GAAATCAC <u>T</u> CAACTTGC	6920
Reduced linolenic acid omega-3 fatty acid	TATCCTCCTCTTCTTTGTTTTTTTTGAGTTCTGAGTCACCTATGGCA AGTTGGGTGATTTGAGAATGTGGGCTAAGGCCACTTCCAAGAATC	6921
desaturase Petroselinum crispum Ser7Term	TATGCCAGGCCCAGAAGTGGAGCTTCATG CATGAAGCTCCACTTCTGGGCCTGGCATAGATTCTTGGAAGTGGC CTTAGCCCACATTCTCAAAATCACCCAACTTGCCATAGGTGACTCAG AACTCAAAAAAAAAA	6922
TCA-TGA	GGTGATTT <u>G</u> AGAATGTG	6923
	CACATTCTCAAATCACC	692
Reduced linolenic acid omega-3 fatty acid desaturase Petroselinum crispum Glu8Term GAA-TAA	TCCTCCTCTTCTTTGTTTTTTTGAGTTCTGAGTCACCTATGGCAAG TTGGGTGATTTCATAATGTGGGCTAAGGCCACCTCCAAGAATCTAT	692
		692
	TGATTTCATAATGTGGG	692
	CCCACATT <u>A</u> TGAAATCA	692

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID
	Reduced linolenic acid omega-3 fatty acid desaturase	CTCTTCTTTGTTTTTTTGAGTTCTGAGTCACCTATGGCAAGTTGGG TGATTTCAGAATGAGGGCTAAGGCCACTTCCAAGAATCTATGCCA GGCCCAGAAGTGGAGCTTCATGTTTCAAC	6929
5	Petroselinum crispum Cys9Term TGT-TGA	GTTGAAACATGAAGCTCCACTTCTGGGCCTGGCATAGATTCTTGGA AGTGGCCTTAGCCC <u>T</u> CATTCTGAAATCACCCAACTTGCCATAGGTG ACTCAGAACTCAAAAAAAAACAAAGAAGAG	
		TCAGAATG <u>A</u> GGGCTAAG	6931
		CTTAGCCC <u>T</u> CATTCTGA	6932
	Reduced linolenic acid omega-3 fatty acid desaturase	ATGAAGCAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTA ATGGTTTTCATGCT <u>T</u> AAGAAGAAGAAGAAGAAGAGGATTTCGACTT AAGCAATCCTCCCATTCAATATTGGTC	6933
10	Vernicia fordii Lys21Term AAA-TAA	GACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATCCTCTTC TTCTTCTTCTTAAGCATGAAAACCATTAACGCCATTTAGAATTG GGGTGTCTTTGTACTGTTGCTGCTTCAT	6934
		TTCATGCT <u>T</u> AAGAAGAA	6935
		TTCTTCTTAAGCATGAA	6936
15	Reduced linolenic acid omega-3 fatty acid desaturase	AAGCAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATG GTTTTCATGCTAAA <u>T</u> AAGAAGAAGAAGAAGAGGATTTCGACTTAAG CAATCCTCCTCCATTCAATATTGGTCAGA	6937
	Vernicia fordii Glu22Term GAA-TAA	TCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATCCTC TTCTTCTTCTTATTTAGCATGAAAACCATTAACGCCATTTAGAA TTGGGGTGTCTTTGTACTGTTGCTGCTT	6938
		ATGCTAAA <u>T</u> AAGAAGAA	6939
		TTCTTCTT <u>A</u> TTTAGCAT	6940
20	Reduced linolenic acid omega-3 fatty acid desaturase	CAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATGGTT TTCATGCTAAAGAA <u>T</u> AAGAAGAAGAAGAGGATTTCGACTTAAGCAA TCCTCCTCCATTCAATATTGGTCAGATCC	6941
	Vemicia fordii Glu23Term GAA-TAA	GGATCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATC CTCTTCTTCTTTATTCTTTAGCATGAAAACCATTAACGCCATTTA GAATTGGGGTGTCTTTGTACTGTTGCTG	6942
		CTAAAGAA <u>T</u> AAGAAGAA	6943
		TTCTTCTT <u>A</u> TTCTTTAG	6944
25	Reduced linolenic acid omega-3 fatty acid desaturase	CAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATGGTT TTCATGCTAAAGAATAAGAAGAAGAAGAAGATTTCGACTTAAGCAA TCCTCCTCCATTCAATATTGGTCAGATCC	6945
30	Vernicia fordii Glu24Term GAA-TAA	GGATCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATC CTCTTCTTCTTATTCTTTAGCATGAAAACCATTAACGCCATTTA GAATTGGGGTGTCTTTGTACTGTTGCTG	6946

Phenotype, Gene, Plant & Targeted	Altering Oligos	SEQ ID NO:
Alteration	CTAAAGAA <u>T</u> AAGAAGAA	6947
	TTCTTCTTATTCTTTAG	6948
	GGTCCAAGCACAGCCTCTACAACATGTTGGTAATGGTGCAGGGAA	6949
educed linolenic acid mega-3 fatty acid	AGAAGATCAAGCTTAGTTTGATCCAAGTGC1CCACCACCC11CAAG	
esaturase	ATTGCAAATATCAGAGCAGCAATTCCAAAA	6950
Slycine max	TTTTGGAATTGCTGCTCTGATATTTGCAATCTTGAAGGGTGGTGGA	0500
yr21Term	GCACTTGGATCAAACTAAGCTTGATCTTCTTTCCCTGCACCATTAC	
AT-TAG	CAACATGTTGTAGAGGCTGTGCTTGGACC	6951
	CAAGCTTA <u>G</u> TTTGATCC	6952
	GGATCAAA <u>C</u> TAAGCTTG	
Reduced linolenic acid	GGTAATGGTGCAGGGAAAGAAGATCAAGCTTATTTTGATCCAAGTG CTCCACCACCCTTCTAGATTGCAAATATCAGAGCAGCAATTCCAAA	6953
omega-3 fatty acid	LACATTOCTCCCACACACACACATTGAGAL I	
desaturase	ATOTOMATCTCTTCTCCCAGCAATGTTTTGGAATIGUIGUIU	6954
Glycine max	GATATTTGCAATCTAGAAGGGTGGTGGAGCACTTGGATCAAAATAA	
Lys31Term AAG-TAG	GCTTGATCTTCCCTGCACCATTACC	
AAG-TAG	CACCCTTCTAGATTGCA	6955
	TGCAATCT A GAAGGGTG	6956
Reduced linolenic acid	TAAACAACATCAAGCTTATTTTGATCCAAGTGCTCCACCACCCTTCA	6957
omega-3 fatty acid	AGATTGCAAATATCTGAGCAGCAATTCCAAAACATTGCTGGGAGAA	
desaturase	LONGOCATTCACATCTCTGAGTTATGTTC	2056
Glycine max	CAACATAACTCAGAGATCTCAATGTGTTCTTCTCCCAGCAATGTTT	6958
Arg36Term	IGGAATTGCTGCTC A GATATTTGCAATCTTGAAGGGTGGTGGAGCA	
AGA-TGA	CTTGGATCAAAATAAGCTTGATCTTCTTT	6959
	CAAATATC <u>T</u> GAGCAGCA	
	TGCTGCTC <u>A</u> GATATTTG	696
Reduced linolenic acid	TATTTTGATCCAAGTGCTCCACCACCCTTCAAGATTGCAAATATCA	696
omega-3 fatty acid	IGAGCAGCAATTCCATAACATTGCTGGGAGAAGAACACATTGAGATO	
desaturase Glycine max Leu41Term AAA-TAA	TOTOACTTATCTTCTGAGGGATGTGTTGG	696
	CCAACACCTCAGAACATAACTCAGAGATCTCAATGTGTTCTT	1 090
	CTCCCAGCAATGTTATGGAATTGCTGCTCTGATATTTGCAATCTTG	-
	AAGGGTGGTGGAGCACTTGGATCAAAATA	696
	CAATTCCA <u>T</u> AACATTGC	
	GCAATGTT <u>A</u> TGGAATTG	696

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- 200 -

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ IE
Reduced linolenic acid omega-3 fatty acid desaturase	CATCCACCGCACCGCACCGCCCGCTGACGGCGCAATGGC CCGGCTCGTGCTCCTAGTGCTCGGGCCTCGCGCCCGCC GCCTGCGCCCGGCCGGGGCGCCATTGCGGCGC	6965
<i>Zea mays</i> Glu8Term GAG-TAG	GCGCCGCAATGGCGCCCCGGCCGGCGCGCGGGCGACGG GCGCGAGGCCCGAGCACT <u>A</u> GGAGAGCACGAGCCGGGCCATTGC CGCCGTCAGCGGGCCGGTGCGGGTGCGGTGGATG	6966
	TGCTCTCC <u>T</u> AGTGCTCG	6967
	CGAGCACT <u>A</u> GGAGAGCA	6968
Reduced linolenic acid omega-3 fatty acid desaturase	ACCCGCACCGCACCGCCCCGCTGACGGCGCAATGGCCCGG CTCGTGCTCTCCGAGTGATCGGGCCCTCGCGCCCGCCT GCGCGCCGGCCGGGCGCCATTGCGGCGCGGTCA	6969
Zea mays Cys9Term TGC-TGA	TGACCGCGCAATGGCGCCCCGGCCGCGCAGGCGGCGCGACGCCGGACGCCGATCACTCGGAGAGCACGAGCCGGGCCATTGCCGCCGTCAGCGGGGCGGGTGCGGGTGCGGGT	6970
	TCCGAGTG <u>A</u> TCGGGCCT	6971
	AGGCCCGA <u>T</u> CACTCGGA	6972
Reduced linolenic acid omega-3 fatty acid desaturase	CCGCACCCGCCCCCGCTGACGGCGCAATGGCCCGGCT CGTGCTCTCCGAGTGCT <u>A</u> GGGCCTCGCGCCGCCTGC GCGCCGGCCGGGCGCCATTGCGGCGCGGTCACC	6973
<i>Zea mays</i> Ser10Term TCG-TAG	GGTGACCGCGCAATGGCGCCCCGGCCGCGCGCAGGCGGCGGACGGGCGAGGCCCTAGCACTCGGAGAGCACGAGCCGGGCCATTGCCGCCGTCAGCGGGGGGGG	6974
	CGAGTGCT <u>A</u> GGGCCTCG	6975
	CGAGGCCC <u>T</u> AGCACTCG	6976
Reduced linolenic acid omega-3 fatty acid desaturase	GCTCGGGCCTCGCGCCGCCGGCCGGGGGCCGGGGCCGGGCCGGCCGCC	6977
<i>Zea mays</i> Ser29Term TCA-TGA	TCGCGGTGGATGGACGCGGACGGCGCGCCGCCGCCGCGGGGGGGG	6978
	GGCGCGGT <u>G</u> ACCCCCCG	6979
	CGGGGGT <u>C</u> ACCGCGCC	6980
Reduced linolenic acid omega-3 fatty acid desaturase	CCCCTCCCCACGCACACGCACAGATCCATCCGCGGCCATGGC CCCCGCAATGAGGCCGTAGCAGGAGGCGAGCTGCAAGGCCACC GAGGACCACCGCTCCGAGTTCGACGCCGCCAAGC	6981
<i>Triticum aestivum</i> Glu8Term GAG-TAG	GCTTGGCGGCGTCGAACTCGGAGCGGTGGTCCTCGGTGGCCTTG CAGCTCGCCTCCTGCTACGGCCTCATTGCGGGGGCCATGGCCGC GGATGGATCTGTGCGTGCGTGGGGGGGGGG	6982

Phenotype, Gene, Plant & Targeted	Altering Oligos S	
Alteration	TGAGGCCG <u>T</u> AGCAGGAG	6983
L	CTCCTGCTACGGCCTCA	6984
	CCTCCCCACGCACACGCACAGATCCATCCGCGGCCATGGCCCC CGCAATGAGGCCGGAGTAGGAGGCGAGCTGCAAGGCCACCGAG GACCACCGCTCCGAGTTCGACGCCGCCAAGCCGC	
	GCGCTTGGCGCGCTCGAGTTCGACCGCGCGTGGTCCTCGGTGGC CTTGCAGCTCGCCTCCTACTCCGGCCTCATTGCGGGGGCCATGG CCGCGGATGGATCTGTGCGTGTGCGTGGGGGAGG	6986
	GGCCGGAGTAGGAGGCG	6987
	CGCCTCCTACTCCGGCC	6988
Reduced linolenic acid omega-3 fatty acid desaturase Triticum aestivum Glu10Term GAG-TAG	CCCCACGCACACGCACAGATCCATCCGCGGCCATGGCCCCCGC AATGAGGCCGGAGCAGTAGGCGAGCTGCAAGGCCACCGAGGACC ACCGCTCCGAGTTCGACGCCGCCAAGCCGCCGC	6989
	GCGGCGGCTTGGCGGCGTCGAACTCGGAGCGGTGGTCCTCGGT GGCCTTGCAGCTCGCCTACTGCTCCGGCCTCATTGCGGGGGCCA TGGCCGCGGATGGATCTGTGCGTGTGCGTGGGGG	6990
	CGGAGCAGTAGGCGAGC	6991
	GCTCGCCTACTGCTCCG	6992
Reduced linolenic acid omega-3 fatty acid	ACGCACAGATCCATCCGCGGCCATGGCCCCGCAATGAGGCCGG AGCAGGAGGCGAGCTGAAAGGCCACCGAGGACCACCGCTCCGA GTTCGACGCCGCCAAGCCGCCCCCTTCCGCATC	6993
desaturase Triticum aestivum Cys13Term TGC-TGA	GATGCGAGGCGCCAAGCCGCCGCCTTGCGCAACTCGGAGCGG GATGCGGAAGGGCGGCGTTGGCGGCGTCGAACTCGGAGCGG TGGTCCTCGGTGGCCTTTCAGCTCGCCTCCTGCTCCGGCCTCATT GCGGGGGCCATGGCCGCGGATGGATCTGTGCGT	6994
	GCGAGCTG <u>A</u> AAGGCCAC	699
	GTGGCCTT <u>T</u> CAGCTCGC	699
omega-3 fatty acid	CTTCACAAATCACAAATCGGAATCAGATCCACCACGACACCCCGG CGGCAATGGCGGCGTAGGCGACCCAGGAGGCCGACTGCAAGGC TTCCGAGGACGCCCGTCTCTTCTTCGACGCCGC	699
desaturase Oryza sativa Ser4Term TCG-TAG	GCGCGTCGAAGAAGAGACGGGCGTCCTCGGAAGCCTTGCAGTC GGCCTCCTGGGTCGCCTACGCCGCCATTGCCGCCGGGGTGTCGT GGTGGATCTGATTCCGATTTGTGATTTGTGAAG	699
	GGCGGCGT <u>A</u> GGCGACCC	
	GGGTCGCCTACGCCGCC	700

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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Reduced linolenic acid omega-3 fatty acid desaturase	ATCACAAATCGGAATCAGATCCACCACGACACCCCGGCGCAATG GCGGCGTCGGCGACCTAGGAGGCCGACTGCAAGGCTTCCGAGG ACGCCCGTCTCTTCTTCGACGCCCCAAGCCCC	
Oryza sativa Gln7Term CAG-TAG	GGGGCTTGGCGCCGTCGAAGAAGACGGGCCGTCCTCGGAAGC CTTGCAGTCGGCCTCCT <u>A</u> GGTCGCCGACGCCGCCATTGCCGCCG GGGTGTCGTGGATCTGATTCCGATTTGTGAT	7002
	CGGCGACC <u>T</u> AGGAGGCC	7003
	GGCCTCCT <u>A</u> GGTCGCCG	7004
Reduced linolenic acid omega-3 fatty acid desaturase	ACAAATCGGAATCAGATCCACCACGACACCCCGGCGCAATGGC GGCGTCGGCGACCCAGTAGGCCGACTGCAAGGCTTCCGAGGACG CCCGTCTCTTCTTCGACGCCGCCAAGCCCCCGC	7005
O <i>ryza sativa</i> Glu8Term GAG-TAG	GCGGGGCTTGGCGCGTCGAAGAAGAGACGGGCGTCCTCGGA AGCCTTGCAGTCGGCCT <u>A</u> CTGGGTCGCCGACGCCGCCATTGCCG CCGGGGTGTCGTGGTGGATCTGATTCCGATTTGT	7006
	CGACCCAG <u>T</u> AGGCCGAC	7007
	GTCGGCCT <u>A</u> CTGGGTCG	7008
Reduced linolenic acid omega-3 fatty acid desaturase	TCAGATCCACCACGACACCCCGGCGCAATGGCGGCGTCGGCGA CCCAGGAGGCCGACTGAAAGGCTTCCGAGGACGCCCGTCTCTTC TTCGACGCCGCCAAGCCCCCGCCCTTCCGCATC	7009
<i>Oryza sativa</i> Cys10Term TGC-TGA	GATGCGGAAGGCGGGGGCTTGGCGGCGTCGAAGAAGAGACGG GCGTCCTCGGAAGCCTTTCAGTCGGCCTCCTGGGTCGCCGACGC CGCCATTGCCGCCGGGGTGTCGTGGTGGATCTGA	7010
	GCCGACTG <u>A</u> AAGGCTTC	7011
	GAAGCCTT <u>T</u> CAGTCGGC	7012

WHAT IS CLAIMED IS:

- 1. An oligonucleotide for targeted alteration of genetic sequence, comprising a single-stranded oligonucleotide having a DNA domain, said DNA domain having at least one mismatch with respect to the genetic sequence to be altered, and further comprising chemical modifications of the oligonucleotide, said chemical modifications selected from the group consisting of an o-methyl modification, an LNA modification including LNA derivatives and analogs, two or more phosphorothioate linkages on a terminus, and a combination of any two or more of these modifications.
- 2. The oligonucleotide according to claim one that comprises two or more phosphorothicate linkages on at least the 3' terminus.
 - 3. The oligonucleotide according to claim one that comprises a 2'-O-methyl analog.
- 4. The oligonucleotide according to claim one that comprises an LNA nucleotide, including an LNA derivative or analog.
- 5. The oligonucleotide according to claim one that comprises a combination of at least two modifications selected from the group of a phosphorothioate linkage, a 2'-O-methyl analog, a locked nucleotide analog and a ribonucleotide.
- 6. The oligonucleotide according to any one of claims 1 to 5 that comprises at least one unmodified ribonucleotide.
- 7. The oligonucleotide according to any one of claims 1 to 6, wherein the sequence of said oligonucleotide is selected from the group consisting of SEQ ID NOS: 4341-7012.
- 8. A method of targeted alteration of genetic material, comprising combining the target genetic material with an oligonucleotide according to any one of claims 1 to 7 in the presence of purified proteins.

- 9. A method of targeted alteration of genetic material, comprising administering to a cell extract an oligonucleotide of any one of claims 1 to 7.
- 10. A method of targeted alteration of genetic material, comprising administering to a cell an oligonucleotide of any one of claims 1 to 7.
- 11. A method of targeted alteration of genetic sequence in callus, comprising administering to the callus an oligonucleotide of any one of claims 1 to 7.
- 12. A method of targeted alteration of genetic sequence, comprising combining target genetic material with an oligonucleotide according to any one of claims 1 to 7, said target genetic material being a non-transcribed DNA strand of a duplex DNA.
 - 13. The genetic material obtained by any one of the methods of claim 8, 9 or claim 10.
 - 14. A cell comprising the genetic material of claim 13.
 - 15. A plant organism comprising the cell according to claim 14.
 - 16. A plant or plant part produced by the method of claim 11.
- 17. A method of determining whether an oligonucleotide is optimized for targeted alteration of a genetic sequence, which comprises:
- (a) comparing the efficiency of alteration of a targeted genetic sequence by an oligonucleotide of any one of claims 1 to 7 with the efficiency of alteration of the same targeted genetic sequence by a second oligonucleotide, said second oligonucleotide selected from the group of an oligonucleotide that lacks the mismatch, a fully modified phosphorothiolated oligonucleotide, a fully modified 2'-O-methylated oligonucleotide and a chimeric double-stranded double hairpin containing RNA and DNA nucleotides.

- 18. The method of claim 17 in which the alteration is produced in a plant cell extract.
- 19. The method of claim 17 in which the alteration is produced in a cell.
- 20. A kit comprising the oligonucleotide according to any one of claims 1 to 7 and a second oligonucleotide selected from the group of an oligonucleotide that lacks the mismatch, a fully modified phosphorothiolated oligonucleotide, a fully modified 2-O-methylated oligonucleotide and a chimeric double stranded double hairpin containing RNA and DNA nucleotides.

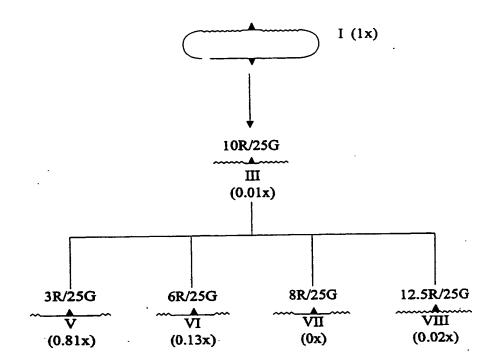


Figure 1A

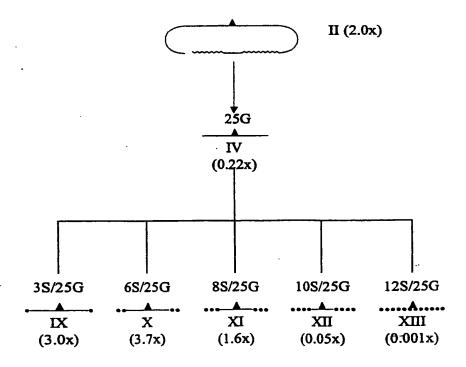


Figure 1B SUBSTITUTE SHEET (RULE 26)

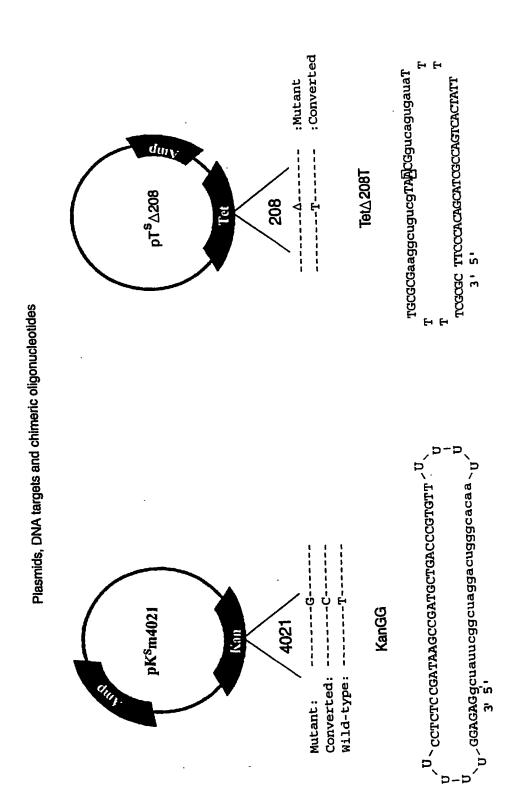


Figure 1C

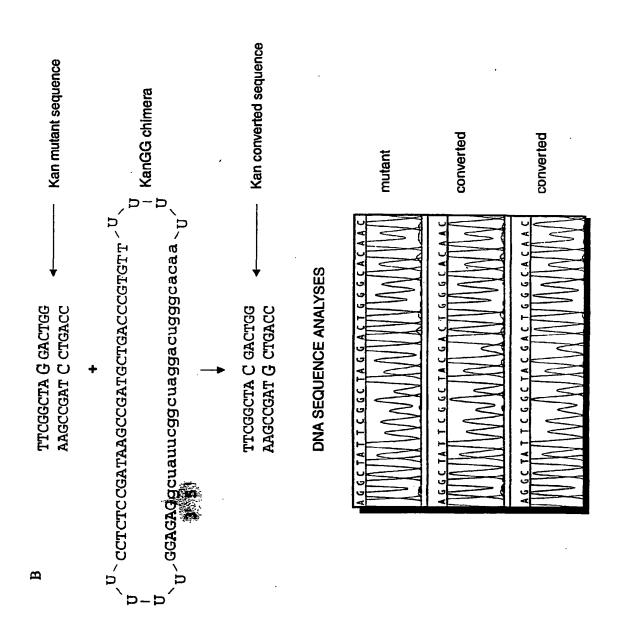


Figure 1D

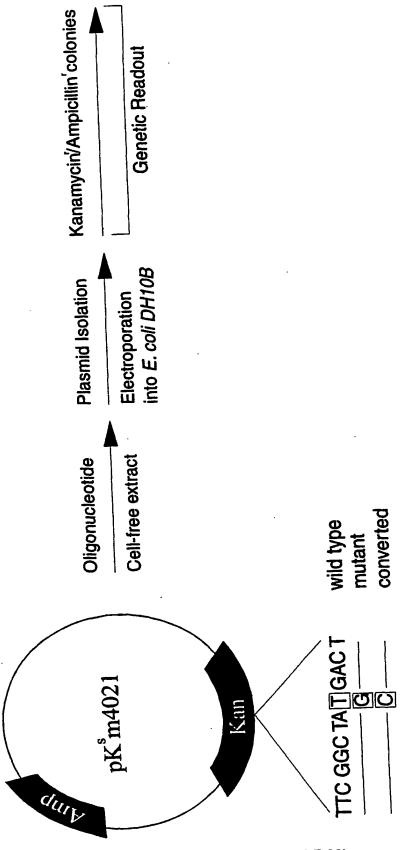
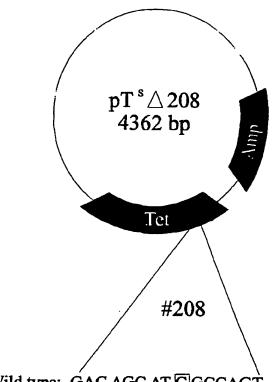


Figure 2

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5/13



Wild type: GAC AGC AT CGCCAGT
Mutant: GAC AGC AT GCCAGT
Converted: GAC AGC AT GCCAGT

Sequence analysis of Tet^r plasmid △208

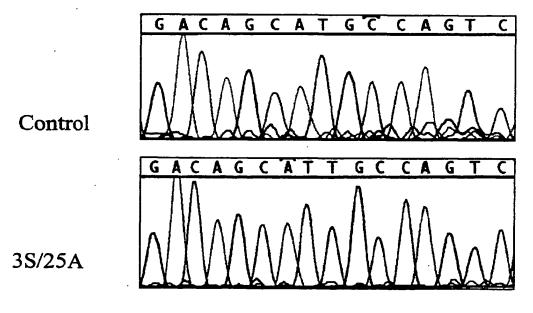


Figure 3

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DNA sequence analysis of Kan^r plasmids

Target codon distribution								
oligomer	TAG	TAC	TAC/TAG	TGG	TCG			
1) 3S/25G (20)		+						
2) 6S/25G (20)		+						
3) 8S/25G (20)		+		 -				
4) 10S/25G (18)		+		+(2)	+(2)			
5) 25S/25G (4)			+(2)	+(2)				

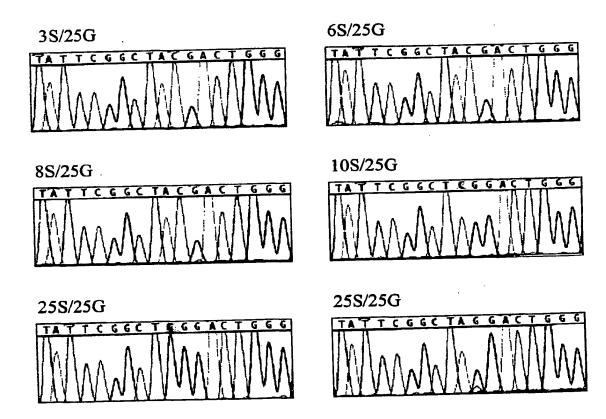


Figure 4

WO 01/92512 PCT/US01/17672

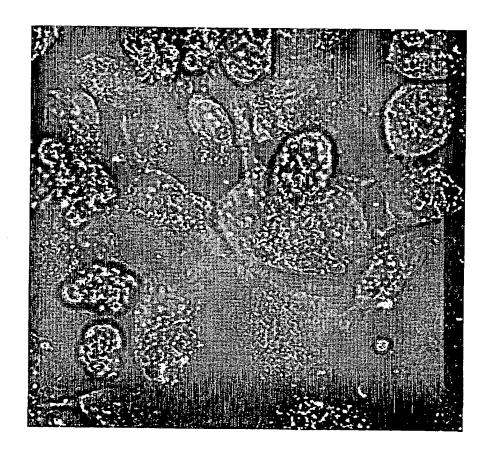


Figure 5

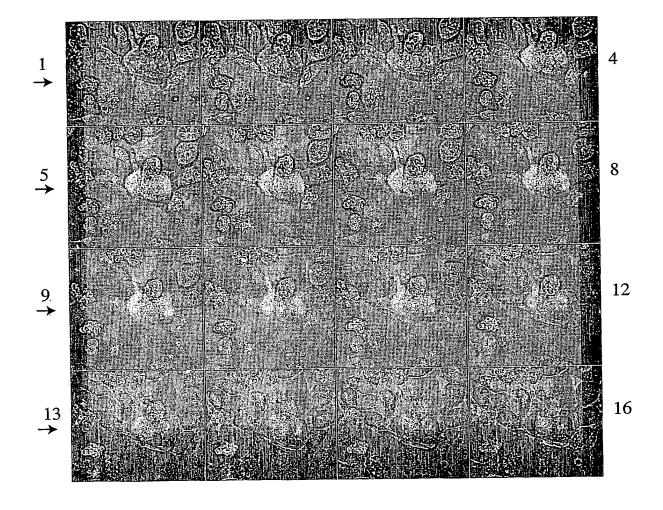


Figure 6

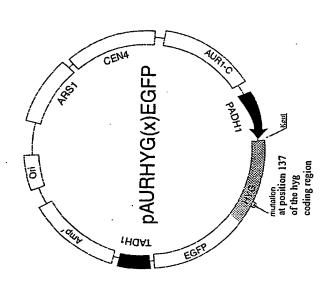
GIGGATAAIGICCI GIGGATAIGICCT Sequence of normal allele: Target/existing mutant: Desired alteration:

GIGGATACGICCI

Figure

Sequence of normal allele: GTGGATATGTCCT GIGGATAGGICCT GTGGATACGTCCT Target/existing mutant: Desired alteration:

Figure 7B



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HygE3T/25: 5'-AGG GCG TGG ATA CGT CCT GCG GGT A-3'

HygE3T/74: 5'-CTC GTG CTT TCA GCT TCG ATG TAG GAG GGC GTG GAT ACG TCC TGC GGG TAA ATA GCT GCG CCG ATG GTT TCT AC-3'

 $\frac{\text{HygE3T}/74\alpha:}{\text{CAG}}$ 5'-GTA GAA ACC ATC GGC GCA GCT ATT TAC CCG CAG GAC GAC GTA TCC ACG CCC TCC TAC ATC GAA GCT GAA AGC ACG AG-3'

HygGG/Rev:

T

T

ACATCCTCCCGCACCTATGCAGGACGCCCAT

T

T
TGTAGGagggcguggaTAGGTccugcgggua
T

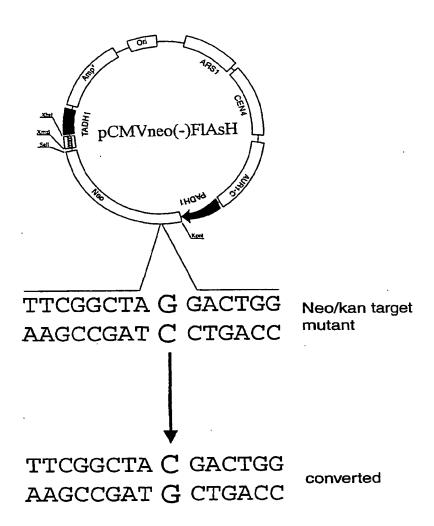
A 3' 5'

T

Kan70T: 5'-CAT CAG AGC AGC CAA TTG TCT GTT GTG CCC AGT CGT AGC CGA ATA GCC TCT CCA CCC AAG CGG CCG GAG A-3'

Figure 8

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FUSION GENE FOR LIGAND BINDING

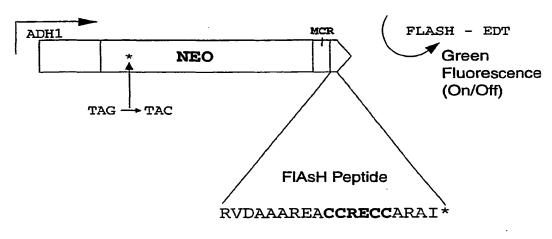


Figure 9
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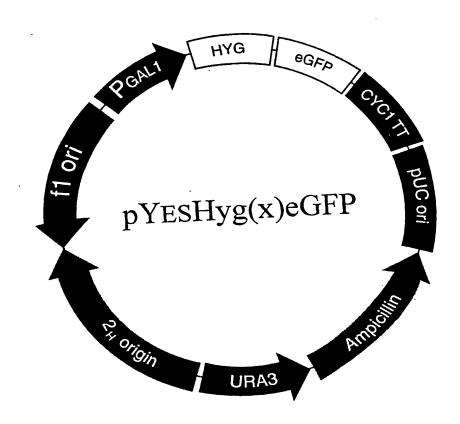


Figure 10

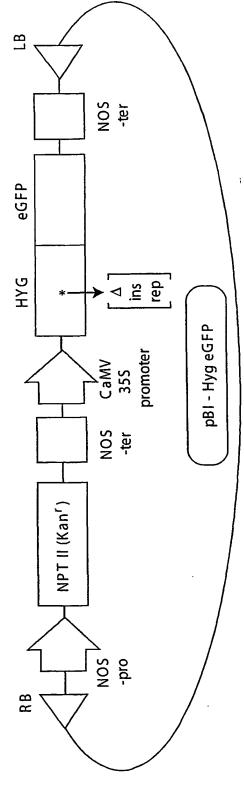


Figure 11

SUBSTITUTE SHEET (RULE 26)

(19) World Intellectual Property Organization International Bureau



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(43) International Publication Date 6 December 2001 (06.12.2001)

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(10) International Publication Number WO 01/092512 A3

(51) International Patent Classification⁷: C12N 15/10, 15/82, 15/11, C07H 21/04, A61K 31/7088, C12N 5/04, A01H 5/00

(21) International Application Number: PCT/US01/17672

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(25) Filing Language: English

(26) Publication Language: English

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(71) Applicant (for all designated States except US): UNI-VERSITY OF DELAWARE [US/US]; 210 Hullihen Hall, Newark, DE 19716 (US).

(72) Inventors; and

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- (74) Agents: HALEY, James, F., Jr. et al., Fish & Neave, 1251 Avenue of the Americas, New York, NY 10020 (US).
- (81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.
- (84) Designated States (regional): ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

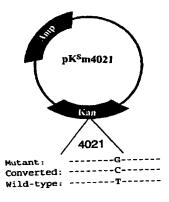
Declarations under Rule 4.17:

as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA,

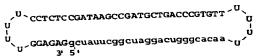
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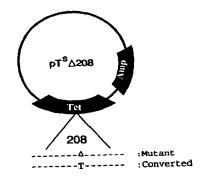
(54) Title: TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES

Plasmids, DNA targets and chimeric oligonucleotides



KanGG





Tet∆208T

TGCGCGaaggcugucgThACGgucagugauaT T TCGGC TTCCCACAGCATCGCCAGTCACTATT

(57) Abstract: Presented are methods and compositions for targeted chromosomal genomic alterations with modified single-stranded oligonucleotides. The oligonucleotides of the invention have modified nuclease-resisant termini comprising LNA, phosphorothioate linkages or 2'-O-Me base analogues or combinations of such modifications.

O 01/092512 A3



CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)

- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)
- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT,

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9 January 2003

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

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A. CLASSIFICATION OF SUBJECT MATTER IPC 7 C12N15/10 C12N15/82 A01H5/00 C12N5/04

C12N15/11

C07H21/04

A61K31/7088

According to International Patent Classification (IPC) or to both national classification and IPC

Minimum documentation searched (classification system followed by classification symbols) IPC 7 C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS, SEQUENCE SEARCH

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° Special	categories of cited documents: "T" la ment defining the general state of the art which is not gidered to be of particular relevance	ter document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention locument of particular relevance; the claimed invention cannot be considered to

Further documents are listed in the continuation of box C.	X Paletti lattilly monitore
 Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filling date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filling date but later than the priority date claimed 	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "&" document member of the same patent family Date of mailing of the international search report
Date of the actual completion of the international search	2 3. 07. 2002
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Name and mailing address of the ISA	Authorized officer
Name and mailing audiess of the low European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	ANDRES S.M.

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International application No. PCT/US 01/17672

INTERNATIONAL SEARCH REPORT

Box i	Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)
This Inte	ernational Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:
1. X	Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely:
	Although claims 8,10,12 (as far as in vivo methods) are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2.	Claims Nos.: because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful international Search can be carried out, specifically:
з. 🗌	Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).
Box II	Observations where unity of invention is lacking (Continuation of item 2 of first sheet)
This Inte	ernational Searching Authority found multiple inventions in this international application, as follows:
	see additional sheet
1.	As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2.	As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3.	As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. X	No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.: 1-20 (all partially)
Remarl	The additional search fees were accompanied by the applicant's protest. No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1. : Claims 1-20 (all partially)

Oligonucleotides characterised by SEQ IDs 4341-4344 for targeted alteration of the Arabidopsis EPSPS at aminoacid position 97; modified forms thereof; compositions and kits comprising them; methods for their optimisation.

Inventions 2. to 668. : Claims 1-20 (all partially)

As for subject 1., but concerning respectively the 667 groups of altering oligonucleotides (SEQ IDs 4345-7012) for each individual mutation disclosed in Tables 10 to 24.

Information on patent family members

International Application No PCT/US 01/17672

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